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# MALIGNANT SYMPATHICUS TUMOR OF THE RIGHT SUPRARENAL *

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Through the work of A. Kohn,[1] Wiesel[2] and others, it is now definitely known that the medullary portion of the suprarenals is of nervous origin. According to Wiesel, certain cells from the neural canal become the building cells of the sympathetic system, and from them arise either chromoblasts, which accumulate in the paraganglia or wander into the suprarenals and later give rise to the chromaffinic cells, or neurocytes, which later develop into ganglion cells. Held[3] states that these cells may also give rise to neuroglia. Now there have been observed tumors partly composed of ganglion cells and nerve fibers, and also new growths containing chromaffinic cells. But the most interesting of all these neurogenic tumors is a very malignant form which occurs apparently only in young children, and is most often found closely associated with the suprarenals. It has long been known that malignant tumors occur in young children in the region of the kidneys, but Wright[4] was the first to describe them accurately and to suggest their close relation to the sympathetic system. Lately Schilder,[5] Landau,[6] and Herxheimer[7] have reported cases of this nature and have discussed them at length. Since these rare new

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1. Kohn, A.: Das chromaffine Gewebe, Ergebn. d. Anatomie u. Entwicklungsgesch, 1902, xii, 253.

2. Wiesel, J.: Beiträge zur Anatomie und Entwicklung der menschlichen Nebenniere, Anatom. Hefte, 1902, xix, 483.

3. Held, H.: Ueber den Bau der Neuroglia, Leipsic, 1903.

4. Wright, J. H.: Neuroeytoma or Neuroblastoma, a Kind of Tumor not Generally Recognized, Jour. Exper. Med., 1910, xii, 556.

5. Schilder, P.: Ueber das maligne Gliom des sympathischen Nervensystems, Frankfurter Ztschr. f. Pathol., 1909, iii, 317.

6. Landau, M.: Die malignen Neuroblastoma des Sympathikus, Frankfurter Ztschr. f. Pathol., 1912, xi, 26.

7. Herxheimer, G.: Ueber Tumoren des Nebennieren-Markes, insbesondere das Neuroblastoma sympathicum, Beitr. z. path. Anat. u. z. allg. Path., 1913, lvii, 112.

growths present phases that are far from being understood at present, I wish to report an interesting instance.

## REPORT OF CASE

*Clinical Diagnosis.*—Sarcoma of kidney; peritonitis.

*History.*—The child, aged 2 years, entered the Kaiser Franz Josef Spital (Professor Bayer's service), April 5, 1914, with the complaint that a swelling of the abdomen had been noted for four weeks. During that time there had been fever. The child had lost flesh, had been peevish, and toward the last had had frequent stools and urination. The father and mother are alive and well; two other children are alive and well, three are dead: one at the age of 7 years, one at 8 months, one at 3 days. There had been no stillbirths.

*Physical Examination.*—This reveals an undersized, anemic child with a hydrocephalic head. The thorax is normal in shape, and the thoracic organs are unchanged. The liver extends over two fingers' breadths below the costal arch on the right side. The liver edge is somewhat irregular. The spleen is not palpable. No fluid can be demonstrated in the peritoneal cavity. There is a firm swelling in the right abdomen between the liver and the crest of the ilium.

*Operation.*—The boy was operated on under ether anesthesia, April 10. A median abdominal incision was made, and a tumor the size of a child's head was found occupying practically the entire right abdominal cavity below the liver. The diagnosis of inoperable sarcoma of the right kidney was made, a drain was inserted, and the wound closed. Death occurred April 18, 1914.

*Necropsy,* eight hours after death (Dr. B. Roman).—The body is that of a male child, 88 cm. long, of slender physique and poorly developed musculature. The subcutaneous fat is sparse. The skin is brownish gray. Post-mortem rigidity is absent. The hair is thin and of a dark blond color. The pupils are narrow, the left slightly wider than the right. The teeth are in good condition. The neck is narrow. The lower part of the thorax is widened on account of the distention of the abdomen. The abdomen protrudes, especially in the right hypogastrium. In the midline above the navel there is an operation wound 10 cm. long, through the lower end of which a gauze drain protrudes. Only the right testicle is in the scrotum. The lower extremities are slightly edematous.

The skull is 51 cm. in circumference, and measures 17.5 by 11.3 by 0.2 cm. The sinuses of the dura contain liquid blood. The parietal bones on both sides are prominent. Their inner surfaces are smooth. The dura mater is adherent to the bones at the vertex of the brain. The leptomeninx is granular and rather moist. The brain weighs 1,370 gm. Its substance is pale and somewhat moist. The ventricles are slightly enlarged. The ependyma is smooth. The hypophysis is slightly enlarged. The surface made by section is of normal color and configuration.

The thyroid gland is somewhat enlarged, yellowish in color, and of normal consistency.

The submaxillary lymph-nodes are enlarged to the size of hazelnuts. Their cut surfaces show reddish and grayish mottlings. The cervical lymph-nodes on both sides are of pea size, of reddish-gray color, and rather soft. The lymph-nodes in the left angulus venosus are enlarged to the size of beans. One of these contains a necrotic area 5 mm. in diameter. The others are similar to the cervical nodes. Those on the right side are unchanged. The peritracheal lymph-nodes are enlarged to the size of cherries. They are grayish red and succulent. The upper right tracheobronchial glands are enlarged, and one of them has a yellowish nodule at its upper pole. The left upper tracheobronchial nodes are like the paratracheal on this side. The lower tracheobronchial nodes

have grown together to form a packet about the size of a small walnut. On the surface made by section these are seen to be composed of grayish-red nodules. The largest of these is 0.7 cm. in diameter. The bronchopulmonary lymph-nodes on the right side are about the size of hemp seeds. They are gray and succulent. The interlobar-pulmonary lymph-nodes on the right side are of pea size. Several of them have grayish nodules near their periphery. Their fellows on the left side are similar in size, but free from nodules. The retromediastinal lymph-nodes form a packet about the size of a date. Cross-section reveals necrotic yellowish-gray nodules, the largest being about the size of a pea.

The thymus is bilobar; each lobe measures 2 by 1.6 cm., and extends from the lower pole of the thyroid gland to the base of the heart.

The pericardial cavity contains a small amount of a clear serous fluid. The pericardium is smooth. The heart is slightly enlarged. The myocardium is fairly firm and of a grayish-brown color. The left ventricle is somewhat dilated. The valves are unchanged. The foramen ovale is patent. The intima of the aorta is smooth.

The right pleural cavity contains a small amount of serofibrinous fluid. The right lung is free. The pleura on the base of this lung is covered with fibrinous exudate and contains a number of ecchymotic spots. The lung is rather bulky, and in the lower lobe its tissue is partly collapsed. The lung crepitates throughout. The upper lobe of the left lung is similar to that of the right, but the lower lobe does not crepitate and contains numerous pneumonic foci. The pleura of this lung is smooth. A purulent fluid can be expressed from the smaller bronchi of this lung.

The mucous membrane of the mouth is pale. The tonsils are the size of beans, and have enlarged lacunae. From one of these on the left side, pus is expressed.

The mucous membrane of the pharynx and of the esophagus is pale.

The peritoneal cavity contains about 100 c.c. of a purulent hemorrhagic fluid. The parietal and visceral peritoneum is covered with a purulent exudate. That part of it which covers the region around the hepatic flexure and the right kidney is very hemorrhagic. The loops of the small intestines are adherent to each other by a fibrinous exudate.

From the region of the right kidney a rather firm tumor protrudes. This has pushed the hepatic flexure forward and upward, and the adherent loops of intestine upward and to the left. The mass extends about five fingers' breadths under the costal arch. It is bounded above by the liver and the pyloric part of the stomach, below by the right anterior-superior spine of the ilium, and to the left by the spinal column. The tumor is a soft, grayish-brown spherical mass, measuring 9 cm. in diameter. It is surrounded by a rather thick connective tissue capsule, which is united to the upper pole of the right kidney. This mass with the adherent kidney occupies most of the space of the right half of the abdominal cavity. The cut section of the tumor is mottled, consisting of grayish-yellow, grayish-red, and dark red areas. The right kidney is markedly compressed from above, downward and inward, by the tumor. The right suprarenal cannot be found; it is apparently included in the mass.

The spleen measures 8 by 7 cm. At its upper pole it is covered by a fibrinous exudate. The surface made by section is grayish red. The follicles are not visible.

The stomach contains a moderate amount of a grayish semiliquid. The mucosa of the stomach is pale and partially softened. The duodenum and bile passages are unchanged.

The pancreas is somewhat compressed by the tumor mass. At the end of the pancreatic tail, there is an accessory spleen of about the size of a cherry.

The abdominal aorta is markedly compressed by the greatly enlarged periaortic lymph-nodes. They also compress the lumen of the vena cava to about

half its normal size. The nodes are closely connected to the tumor by connective fibers and are diffusely infiltrated by the tumor tissue. Similar glands surround and compress the left renal vessels. The lymph-nodes around the head of the pancreas are enlarged to the size of beans, and are grayish and succulent. So, also, are the paragastric lymph-nodes, and the nodes in the hilum of the liver.

The liver extends a hand's breadth below the costal arch. The right lobe of the liver is flattened out and compressed by the new growth. The tumor adheres to it. The capsule of the liver is partially covered with a fibrinous exudate. Numerous tumor masses of various sizes are seen in the liver parenchyma. Some of the external nodules have depressed centers. The remaining liver tissue is pale gray. The lobular markings are fairly distinct.

The left suprarenal is slightly enlarged and flat, and the medullary substance is markedly diminished in amount.

The left kidney measures 7 by 4.5 cm. Its capsule strips easily. Its surface is smooth. The pelvis and the ureter of this kidney are unchanged.

The mucous membrane of the urinary bladder contains numerous pin-point sized hemorrhages near the fundus. The urethra is unchanged. The testicles are small. The rectum is practically empty. The external genitalia are unchanged.

The mesenteric lymph-nodes are moderately enlarged, the largest being bean-sized. These are grayish-red to red in color.

The rest of the alimentary tract is unchanged.

In the body of the fourth thoracic vertebra there is an indefinitely outlined grayish nodule about the size of a pea. A similar, but smaller, nodule surrounded by a hemorrhagic zone occurs in the fifth thoracic vertebra.

The medullary substance of the left femur is dark red with yellowish-red mottlings. In the region of the proximal epiphysis occurs a hemorrhagic area, about 1 cm. in diameter, which at this place fills the entire medullary cavity. The medullary substance of the right femur is similar to that of its fellow, except that the former is more mottled. In the medullary substance of the right tibia, there occurs a grayish nodule the size of a hemp seed. There is a similar nodule in the medullary substance of the right ulna. The medullary substance of the left tibia, right fibula and left ulna are slightly mottled. That of the left fibula is unchanged.

In one of the left ribs there occurs a soft grayish-yellow nodule about 1.5 cm. in length. This entirely fills the medullary cavity of the bone.

*Bacteriologic Examination.*—From the peritoneal exudate, *Staphylococcus albus* was obtained in pure culture.

*Anatomical Diagnosis.*—Fresh laparotomy wound with partial healing. General, acute, seropurulent peritonitis. Serofibrinous pleurisy, right. Malignant tumor of right suprarenal (?) with compression and displacement of kidney and displacement of hepatic flexure. Metastases in the retroperitoneal lymph-nodes with compression and partial stenosis of the vena cava; in the retromediastinal; the right bronchopulmonary, the right lower tracheobronchial, and the left supraclavicular lymph-nodes; in the liver, the thoracic vertebra, and in numerous long bones. Slight hydrothorax with anasarca. Lobular pneumonia of the lower lobe of the left lung. Parenchymatous degeneration of liver, kidneys and of the heart muscle, with slight dilatation of the left ventricle. Catarrhal bronchitis. Old lacunar angina. Slight hydrocephalus. Patent foramen ovale.

*Microscopical Examination.*—Technic: The specimens were hardened in formaldehyd and Zenker's fluid, and embedded in paraffin and celloidin. Frozen sections were cut for Bielschowski's stain. The stains employed were: hemalum and eosin; iron hematoxylin; Van Gieson's stain; Mallory's connective tissue and neuroglia stains; Wiegert's neuroglia stain and also Alzheimer's stain for ameboid neuroglia stain; and finally Bielschowski's stain for nerve fibers.

The largest tumor is surrounded by a rather thick connective-tissue capsule from which trabeculae extend inward to form the framework of the new growth. On the inner surface of the capsule, there occur numerous remnants of the suprarenal cortex. In places these cells extend in narrow columns between the connective tissue fibers of the capsule; in other parts they form distinct nodules, which, in turn, are invaded by the tumor tissue. Most of the cortical cells are atrophic and degenerated. The posterior part of the capsule is free from these cells.

Fig. 1.—Photograph of main tumor and the neighboring organs. The tumor has been cut into halves and the surface made by section shows large areas of necrosis and numerous hemorrhages. The right kidney is compressed by the new growth. The liver is seen studded with tumor nodules.

The great bulk of the new growth itself is composed of cells averaging 8.5 microns in diameter. The individual cells are arranged either concentrically around fibrillar centers, or occur diffusely scattered. The concentrically arranged cells, termed by Küster[8] rosettes, form a conspicuous part of the tumor. The rosettes are round to oval, measuring from 26 to 54 microns in one diameter, and from 51 to 79 microns in the other diameter. Their periphery is formed of from twenty to thirty individual cells, which are usually arranged in a single layer. At times a more or less complete double layer is observed.

8. Küster, H.: Ueber Gliome der Nebennieren, Virchows Arch. f. path. Anat., 1905, clxxx, 117.

The nuclei of these cells are slightly oval or round, and measure, in most cases, from 5 to 9 microns. They are rich in chromatin and have a distinct nuclear membrane. The smaller the nucleus, the richer it appears to be in chromatin; thus, the few nuclei present which are less than 5 microns stain very heavily, and the largest have a vesicular appearance. Mitotic figures rarely occur among the cells in the rosettes. The cytoplasm surrounds the nucleus as a delicate, thin zone, massed at the centrally directed end of the cell so as to form a sort of short stalk from which one or more fibrillae run to lose themselves in the central fibrillar structure of the rosette. The entire central structure is thus formed by these fibrils. Smaller cytoplasmic projections extend in other directions from each cell. In certain fields the fibrillar structure is especially marked, each cell sending out from one to eight fibrillae, which form a network. Nowhere in any section is there any tendency of the fibrillae to form parallel bundles. In certain portions of the tumors where rosettes are practically absent, the cells are closely packed and few fibrillae are seen. The nuclei of these cells are all more or less oval, and mitotic figures are more frequent than among the cells forming rosettes.

Throughout the tumor, large, oval nuclei, poor in chromatin, occur singly or piled one against the other forming clumps. These clumps consist of from three to ten nuclei, measuring 9 to 12 microns in diameter, and, where the tumor cells are diffuse, they are apparently without any cytoplasm. In most instances, however, these nuclei are connected with an undivided smaller or larger portion of cytoplasm, in which the nuclei sometimes lie near one end, thus simulating giant cells (Fig. 3). Structures of this kind are most numerous where the connective tissue is well developed, or just underneath the capsule where this contains remnants of the suprarenal cortex. Here these multinucleated structures form a large part of the tumor. In some places, the nuclei form a round ring about a center of undivided protoplasm. In other parts, groups of these large nuclei occur at the two poles of cytoplasmic masses. In still other places, one finds large areas of multinucleated cytoplasm, part of which is fibrillar, part homogeneous; and, also, an occasional small heavily stained nucleus definitely connected to the syncytium by fibrillae running directly into the undivided cytoplasm. There also occur two other structures among or near connective-tissue trabeculae and near the capsule of the tumor. One of these is a distinct type of large, often multinucleated cell in which the cytoplasm is abundant, and either round, unipolar, or multipolar. The nuclei are also rather large and have a distinct nuclear membrane and a large nucleolus. These cells are not numerous, are scattered, and show, in every case, distinct evidences of degeneration. Wherever these cells occur there are also found oval or round, hyaline bodies, measuring from 15 to 20 microns in diameter, lying among the connective tissue. These do not take the stain of corpora amylacea. The smaller tumor cells do occur in scattered heaps among these structures, but the syncytial tissue, the large polar cells, as well as the hyaline bodies, are absent in actively growing portions of the tumor.

The fibrillae in the center of the rosettes stain red with eosin, yellow with Van Gieson, but do not stain at all with Mallory's connective tissue stain. With Mallory's neuroglia stain most of the fibrillae take a light blue color; but a few wiry fibrillae, both within the rosettes and outside, stain a heavy blue. Wiegert's neuroglia stain gives a negative result. Alzheimer's stain for ameboid glia cells stains cytoplasm and fibrillae, as well as almost everything else, blue. With Bielschowski's stain the center of the rosettes takes a brownish tinge, but repeated attempts failed to demonstrate any fibrillae which stain black. On the other hand a few distinctly black, rather thick fibers occurred near the large ganglion cells.

The connective tissue is scant in certain places throughout the tumor; in other places it forms over half of the substance. Between these two extremes variations occur. Where the trabeculae are thick, hyaline degeneration is

marked. Throughout the capsule, groups of tumor cells are abundant. They are seen forcing their way, as it were, between the bundles of supporting tissue. The vessels of the capsule are in many places crowded with cells from the new growth.

The blood-vessels enter the tumor through the trabeculae. They are abundant in places; in other portions they are not frequent; in all parts they are thin-walled and have wide lumina. Arteries cannot be recognized. The blood contains an unusually large proportion of polynuclear leukocytes. Smaller and larger hemorrhages are noted throughout, but in greatest abundance in what appears to be the older parts of the tumor. Some of them are recent; others seem old; a few are calcified. Frequently, the hemorrhages are associated with the areas of necrosis of the tumor cells. Such areas are also rather numerous so that some sections are composed almost entirely of necrotic cells. Then

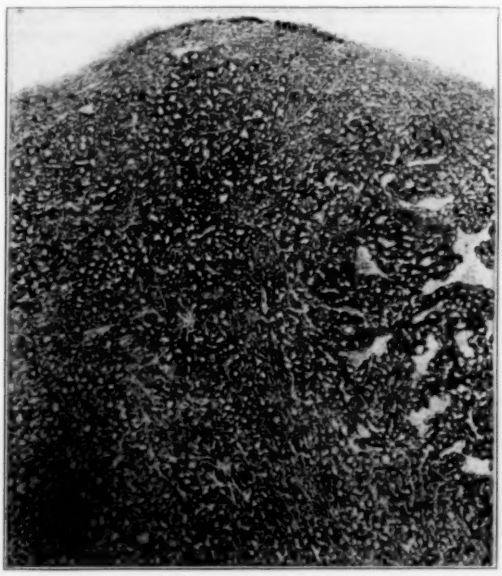

Fig. 2.—Photomicrograph (retouched, × 25) of area from main tumor. The rosettes are very abundant, and the blood-vessels have wide lumina.

again, they are practically absent in others. All stages, from degenerated, poorly staining cells to complete coagulation necrosis and even calcification, occur.

The microscopic structure of the large nodules and of two of the smaller ones lying next to the main tumor is very similar to that described. The connective tissue is rather abundant, and among the connective tissue fibers are seen degenerated ganglion cells, and also single or grouped polygonal cells with large nuclei — evidently chromaffinic cells. Rosettes are in some parts abundant. In the smallest of the three nodules, the cells are in general diffusely arranged, and mitotic figures are abundant. In certain fields of the large nodules, the fibrillae are numerous. Hemorrhages are frequent in all three. Large portions of the nodules are necrotic. Tumor cells occur in the lumina of many of the blood-vessels.

The remaining nodules in the abdomen are infiltrated lymph-nodes. Ganglion and chromaffinic cells are absent, the nodules being composed entirely of well-preserved or necrotic tumor cells. In these nodules, many lymph-vessels are filled with tumor cells.

The right lobe of the liver is markedly infiltrated with cells from the new growth. In the extreme right edge of the liver, necrosis is prominent among the tumor cells, but nearer the left lobe nodules of various sizes occur, which afford an interesting picture (Fig. 4). In these smaller nodules, the liver cells in places have been totally destroyed, so that only an occasional bile-duct is seen. Toward the periphery occur atrophic columns of liver cells, forming a sort of scaffold for the proliferating tumor cells. Rosettes are abundant and similar in size and structure to those already described except that a larger number of them have a double row of cells. Mitotic figures are rather frequent even among the nuclei of the rosette cells, and are very abundant in the cells outside the rosettes. The nuclei are somewhat irregular in size. The small, heavily stained ones are numerous, and many large (10 microns), round vesicular nuclei also occur. Here, as in the rest of the metastases, clumps of large, oval nuclei are scattered among the rosettes. Gland-like arrangements occur in some places. At the periphery of the nodules the tumor cells extend through the liver sinusoids. These are crowded with cells, and cell division is frequent. Thrombi consisting of tumor cells also occur in the portal veins. Along the periphery of the liver, just under and in the capsule, there is a practically continuous zone of tumor cells. About half of these are of the smallest variety described. The rest of the liver cells show a marked degree of parenchymatous degeneration.

The enlarged glands in the mediastinum and those around the trachea and bronchi all show metastatic nodules, but most of them are very necrotic. In these glands, rosettes and fibrillae are comparatively few.

Practically all of the long bones of the body show tumor metastases in the medullary substance. The structure is characteristic; hemorrhages occur, the fibrillae are exceedingly well developed, and the rosettes are abundant (Fig. 5). Here as in the other metastases, ganglion and chromaffinic cells are absent.

The lungs are free from metastases. The rest of the organs show no interesting changes.

## COMMENT

The striking features of this new growth then are several. In the first place, it has all the earmarks of a malignant growth; for, on the one hand, its growth is infiltrative, as shown by the invasion of the suprarenal cortex by the tumor cells, by the cells breaking through the capsule and then forming nodules outside of it, and by the infiltration of the connective tissue of the liver. This infiltrative nature is distinct, but not so pronounced as in certain other tumors. Thus, it seems to have a certain respect for the organs, since nowhere have the tumor cells invaded the compressed kidney or any of the large blood-vessels whose lumina it had narrowed by its expansive growth. This respect for organs on the part of the tumor cells has been observed in other cases of this type. Both Landau and Herxheimer have called attention to it. On the other hand, true metastases occur. Pick and Bielschowski[9] have suggested a multiple origin for new growths of this

9. Pick, L., and Bielschowski: Ztschr. f. d. ges. Neurol. u. Psychiat., 1911, vi, No. 4.

type. There can be no question but that the growths in the lymph-nodes and other organs, in this case, are secondary. Since the tumor cells are observed in numerous places both in the blood- and the lymph-vessels, it is evident that the elements of the new growth were distributed by means of both these channels. The tumor cells must first have reached the neighboring lymph-nodes, since the nodules here are the largest and show the most extensive necrotic areas. From these nodes the cells reached the main lymph trunks of the abdomen and extended upward, producing metastases in the nodules in the hepatic hilum, and from there entered the liver. It is also evident that the

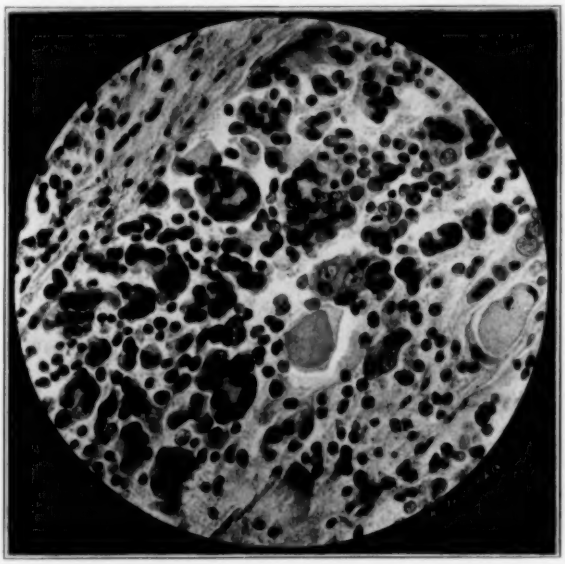

Fig. 3.—Photomicrograph (retouched, × 370) of area near capsule of the main tumor, showing multinucleated masses of cytoplasm and degenerated ganglia cells.

liver received implantations through the blood-stream. Following the lymph-stream further we see that the malignant cells involved the glands in the mediastinum, those around the bronchi and those in the neck. The cells, then, reached the venous circulation, first through invading the vessels of the new growth; second, by the thoracic duct, and thirdly, through the hepatic veins; thus, eventually, tumor cells must have reached the lung in numbers. A careful search of both lungs failed to reveal any nodules, yet the cells must have passed through the lung capillaries to reach the arterial blood in order to produce metas-

tases in the bones, for the multiple metastases in the various bones can be explained in no other way. Just why the bones alone should be involved cannot be accounted for. To-day, we only know that certain malignant tumors, such as neuroglioma of the retina, carcinoma of the prostate and of the thyroid, have a certain affinity for bone. The future must explain why. The most marked metastases occur in the liver. This is the organ most frequently involved in these cases; indeed, in some of the cases reported, the liver is practically the only organ containing metastases. Herxheimer believes that the reason lies in the fact that the liver forms a good medium for the new growth. It is also reasonable to suppose that it receives the greatest number of implantations. The bone metastases are interesting, in the first place, because of the general involvement of the long bones, and, in the second place, because of the beautiful rosette formation and fibrillae in these metastases. Bone metastases have been observed in less than one-half the cases reported. The statement is made by Landau and others that the growth in bone cannot be recognized as sympathicus tumor. In our case, this is certainly not true, as the growth could not be mistaken for anything else. Because of the fact that the tumor tissue is difficult to recognize macroscopically, and, also, because the bone-marrow is rarely examined as a routine, I am inclined to believe that metastases may be more frequent than reports would indicate.

In the second place, this type of new growth apparently occurs only in young children, and most of the reported cases have been in very young children. Indeed, if we except certain dubious tumors, such as the second case of Küster, and two of Wright's cases, the oldest case reported is in a child of 7. Among the twenty-eight cases, reports of which were collected by Herxheimer, twenty were less than 1 year, and twelve had not reached the age of 3 months; so that the case here reported must be considered as occurring in an unusually old child. The early occurrence of these tumors is of course very strong evidence for their congenital origin.

The third peculiarity of this tumor is its location. There can be no reasonable doubt as to the origin of the tumor in this case. Its location and the fact that in the capsule, as well as in the substance of the tumor, practically the entire cortex of the right adrenal was found, and also that throughout the tumor nervous remnants of the medulla occurred, show it to have sprung from the medullary substance of the suprarenal. Most of the tumors of this type have had their origins in the medulla of one of the suprarenals or have sprung from the sympathetic system near the kidneys. And every one has originated in some part of the sympathetic system. The fact that tumors so fre-

quently occur in the suprarenals must be borne in mind by the clinician in the differential diagnosis in this part of the body in the young.

Finally, this tumor has a structure which distinguishes it rather sharply from the other known types of malignant tumors. The rosette that forms such a conspicuous part of the cellular structure of this tumor is probably not identical with any other structure known. These are present in greater or smaller number in practically all tumors of this kind, and the fibrillar substance between the diffusely arranged cells is always present in greater or smaller amount.

The other features of this case, such as the frequent hemorrhages, the large areas of necrosis and the wide blood-vessels with exceedingly

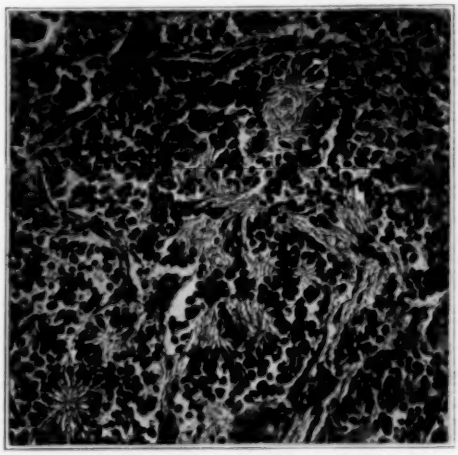

Fig. 4.—Photomicrograph (retouched, × 75) from small nodule in liver. The fibrillae of the rosettes stand out prominently.

frail walls, have been observed in all the other tumors of this type. The microchemical peculiarities, which make the fibrillae stain yellow with Van Gieson and make them fail entirely to react with the connective tissue stain, are also noteworthy. Therefore, the location of the tumor, its structure and the staining reaction of the fibrillae make it certain that the tumor cannot be classed either among the sarcomas or the ordinary carcinomas. Nor does it belong to the new growth of lymphatic tissues, the lymphosarcoma, which it simulates macroscopically, but must arise from neurodermal cells.

All present-day writers agree that these tumors are nervous in origin, but as to the particular nervous element from which they spring, there exist two opinions. One group believes that these tumors

are composed of so-called building cells of the sympathetic system and that the fibrillation observed is an attempt toward differentiation into nerve fibers. In other words, the tumors are what Herxheimer calls neuroblastomas. The other group holds the tumors to be malignant gliomas. Both these views are based, of course, on the resemblance which the various observers see between the elements of these new growths and the normal cells. In order to make the relation of these growths to the nervous elements more clear, a brief statement of the normal development of the sympathetic and the chromaffinic system is offered.

Kohn, Wiesel, Held[10] and others have shown that certain cells from the neural canal become the mother cells of both the sympathetic and the chromaffinic systems. Wiesel holds that some of these building cells wander into the suprarenals to form the medulla, and that these cells (for the most part) give rise to chromaffinic cells, but that from them also spring the ganglion cells and other nervous elements of this organ. But Zuckerkandl[11] holds that the cells which wander into the adrenal cortex later develop only into chromaffinic cells, or, at least only in exceptional cases, give rise to other nervous elements. Held has further shown that, as a first step in differentiation, the sympathetic neurocyte undergoes a fibrillation of its cytoplasm. Finally, Held has shown that glia cells also occur in the sympathetic system, and that this glia tissue differentiates from heaps of large oval nuclei lying in undifferentiated cytoplasm—a true syncytium. It is also to be borne in mind that Kohn has shown that young chromaffinic cells occur in two forms, as a syncytium and as heaps of cells, and that in man the syncytial form predominates, but that the clumps also are found.

Based on this somewhat conflicting knowledge of the embryonal development of the sympathetic system, Wright, Landau, Herxheimer and Martius[12] hold that this type of tumor comes from sympathetic formative cells, and that its cells have a tendency to develop into true nerve cells. The reasons they give for their position are positive as well as negative. In the first place, Wright sees a striking similarity between groups of tumor cells and those of the sympathetic nerve cells in young children. He also finds bundles of fibrillae running parallel to each other in the tumor in very much the same way that the nerve

10. Held, H.: Die Entwicklung des Nervengewebes bei den Wirbeltieren, Leipsic, p. 378.

11. Zuckerkandl, E.: Keibel-Mall, Handbuch der Entwicklungsgeschichte des Menschen, Leipsic, 1910, xi, 157.

12. Martius, K.: Maligner Sympathoblastoma des Halssympathikus, Frankf. Ztschr. f. Pathol., 1913, xii, 442.

fibers do in the young ganglia. Then, he fails to stain the fibrillae with Mallory's neuroglia stain.

Landau has also observed the parallel arrangement of the fibrillae. He further adds that the cells of the new growth are capable of differentiating into higher nervous elements, so that the older the individual, the more differentiated the cells. Thus, ganglion and glia, as well as chromaffinic cells, may appear in this type of tumor when older children are affected. Martius supports Landau's theory by a case of a tumor from the cervical sympathetic in a 2½-year-old child, in whom well-developed ganglion cells occurred. At first glance, it may seem as though our tumor also lends proof to Landau's contention. But the nervous and the chromaffinic elements found in this

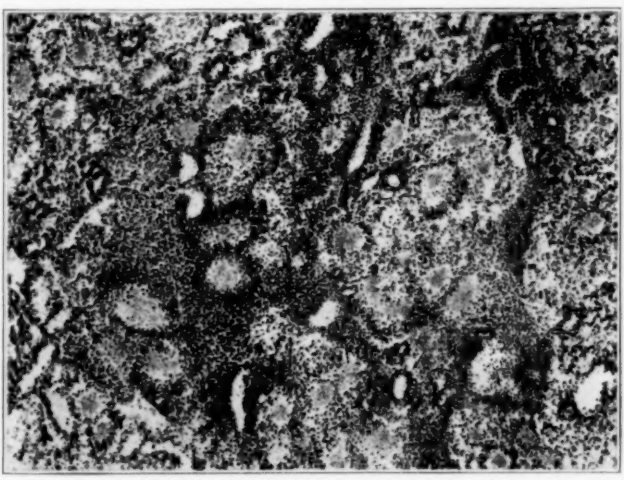

Fig. 5.—Photomicrograph (retouched, × 50) from metastasis in one of the long bones.

case probably do not belong to the blastoma, since they occur only where such tissue normally are found, and in normal amount without sign of proliferation, but rather of marked degeneration. Landau admits the possibility that these tumors may contain neuroglia cells, but does not believe in their gliomatous nature, because the fibrillae do not take the glia stains, because glia cells are higher differentiation products and because the rosettes are widely different from the glia rosettes.

Herxheimer supports the arguments advanced by Wright and Landau. He applied the Bielschowski stain and found that the fibrillae stained black. This he considers conclusive proof that the tumor is a neurocytoma.

The evidence cited is not invulnerable. Thus Wiesel's conclusion that the formative cells of the sympathicus enter the suprarenals is not accepted by all. The similarity between the tumor cells and the young sympathetic ganglia, emphasized by Wright, does not appear striking to me. And it is curious to note that the arrangement of the fibrillae into parallel bundles, noted by Wright and made much of by Landau and Herxheimer, occurred most strikingly in the case of Wright which both Landau and Herxheimer agree does not belong to this type of new growth. It may also be questioned whether the similarity between the fibrillation occurring in sympathetic neurocytes as seen by Held, and the fibrillae of these new growths, should warrant the emphasis laid on it by some. Herxheimer admits that both connective-tissue fibers and nerve fibers stain with Bielschowski. It must also be borne in mind that the section stained is from the medulla of the suprarenal where nerve fibers normally are abundant. He failed to find any nerve fibers in the other metastases. In the case here reported, repeated attempts with Bielschowski stain failed to stain any of the fibrillae black.

Those who consider these tumors malignant gliomas include Virchow,[13] Küster, Lapointe and Lacene[14] and Schilder. They are less certain in their assertions. Thus, Virchow speaks of the tumors as glia-like, and Küster states that, since they are more like gliomas than any other tumor, he ventures the name "malignant glioma." The main points on which this view is based are, first, that the tumor cells are strikingly like neuroglia cells, and second, that the interlacing fibrillar network in this type of tumor cannot be distinguished from such network found in gliomas. Neither of these points can be gainsaid. These authors also assert that rosettes occur in gliomas. It is true that most of the glia rosettes are not identical with the structures here known as glia balls, spheres or rosettes. Landau's statement that these could not possibly be mistaken for glia rosettes is too strong, for there does occur in the tumors, known as neurogliomas of the retina, rosettes very similar to these.

Schilder rightly calls attention to the fact that unripe glia fibrillae do not take the specific stain for neuroglia; thus, tumors undisputably neurogliomas often fail to take the glia stains. Besides, Weigert's stain is notoriously uncertain even for normal glia fibrillae. In this case, some of the fibrillae in most of the rosettes stained deeply with Mallory's neuroglia stain.

In conclusion, I wish to call attention to the many similarities between this type of new growth and the neuro-epitheliomas or neuro-

13. Virchow, R.: Die krankhaften Geschwülste, 1864, ii, 150.
14. Lacene, cited by Landau (Footnote 6).

gliomas of the retina. Both types occur in the young; both are composed of cells with chromatin-rich nuclei and fibrillar cytoplasm; both have rosettes; both are malignant with tendency toward multiple bone metastases.

It does not seem to me that we have at the present time evidence enough to place this new growth either among the neurocytomas or the neurogliomas. The matter of classification is of minor importance. Certain it is that we are here dealing with a malignant sympathicus tumor.

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# UREMIA

## III. THE NON-PROTEIN NITROGEN OF BLOOD *

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The nitrogen-containing crystalloids of blood have interested physiologists and clinicians for almost a century and in some respects the questions at issue are but little clearer to-day than in the time of Bright. Methods of research have changed and the data secured are more accurate, but it is remarkable in reviewing the literature that many of the conclusions of the later investigations are a simple rewording of the earlier. The facts have, however, been amplified and our knowledge made more complete without as yet any unaminity of opinion as to the significance of these facts. The amount of data at our disposal now is not inconsiderable and it may not be a useless task in the beginning to survey this field.

Urea was first detected in the urine (*matière savonneuse*), by Rouelle, Jr., in 1773, and in 1821 Prevost and Dumas observed an accumulation of urea in the blood of dogs following extirpation of the kidneys. It was this latter observation particularly which led Bright's assistants Prout, Babbington and Christison,[1] to analyze the blood of nephritic patients. From the fact of finding urea in the blood of patients with severe nephritis the name of the syndrome, uremia, took origin. It was not learned till some time later that urea is only slightly toxic. Investigations bearing on the urea content of blood were taken up by German and French clinicians and quantitative determinations attempted. Babbington and Christison had made some estimations of the amount of urea in body fluids. Their method was to desiccate the blood or fluid and then reduce the dry residue to a fine powder which was repeatedly extracted with alcohol. From the alcohol extract, on addition of nitric acid, crystals of ureanitrate separated out and were collected and weighed. By this method 15 grains of urea were recovered from 1,000 grains of blood. Urea was also found in the fluid of the ventricles of the brain. Picard,[2] also using alcohol as a specific urea solvent, investigated the relation that the amount of blood-urea bears to the taking of food. Estimating the urea by Liebig's titration

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1. Bright: Guys Hosp. Rep., 1836, 1, 339.
2. Pickard: Thèse de Strassburg, 1856.

method, he found from 0.05 to 0.07 per cent. during fasting and from 0.11 to 0.13 per cent. after food. The methods at the disposal of investigators at this period were not accurate and the results only approximate. The blood-proteins were either precipitated by alcohol or by means of a salt, such as sodium sulphate, and urea determined by the titration method of Liebig; or by measuring the carbon dioxid, evolved on decomposition (Millon). Discrepancies and variations in normals are to some extent explicable on these grounds. Without entering further into specific details the net result of the earlier investigations indicated that the non-protein nitrogen (since the methods used determined more than urea) varies under normal conditions and is influenced by the ingestion of food; and that it is often above the normal maximum with grave cases of nephritis: 0.15 per cent. (Picard); 0.2 per cent. (Yvon); from 0.28 to 0.41 per cent. (Gréhant). Also that this supernormal increase occurs in some other diseases, notably cholera and typhus fever.

An elaborate reexamination of the whole field was conducted by Schöndorff[3] using methods in some respects more accurate than those of his predecessors. He held a faulty conception of the range of substances precipitated by phosphotungstic acid, which does not, however, seriously detract from the value of his work. The earlier work was confirmed by Schöndorff in so far as his clinical material permitted. From the clinical aspect the important observation was, that of the total non-protein nitrogen in blood, urea composes a variable fraction; in the blood of a dog 79.6 per cent. of the total nitrogen, and in that of a man with dyspnea 56 per cent.

During the period since Schöndorff a considerable amount of careful work has been conducted by methods which, although not always entirely accurate, yet are sufficiently good so that the results may be considered. The chief obstacle in determining non-protein nitrogen in the blood has been the primary separation of the proteins. To effect this end a number of procedures are now in current use and will be reviewed, since I have controlled them by intercomparison, using the Kjeldahl method for nitrogen. The common method of protein separation by coagulation with heat and acid can not be used since protein is somewhat hydrolyzed even on short exposure to heat in acid solutions. As a result the filtrate invariably contains some of the cleavage products—peptones, polypeptids, etc.—which vitiate the accuracy of the final results, even when sodium chlorid is added, as used by Ascoli. The amount of these substances formed is not large enough so that a normal blood might appear pathological; but the results by this method constantly run higher than controls by safer procedures.

3. Schöndorff: Arch. f. d. ges. Physiol. (Pfluger's), 1899, lxxiv, 307.

Coagulation by means of alcohol is one of the oldest methods and is still used. When one volume of blood is added to from eight to ten volumes of 95 per cent. alcohol the filtrate contains no protein. Alcohol is, however, an excellent solvent for lipoids; of these in the blood some, for example, lecithin, contain nitrogen. Determinations by this method also run slightly higher than they should and actually higher than the control determinations by colloidal methods. Either ethyl or methyl alcohol may be employed.

Hohlweg and Meyer devised a method wherein the diluted blood is made acid with dilute acetic acid and 5 per cent. solution of diacid-potassium phosphate and then half saturated with sodium chlorid. The method works well so far as biuret tests indicate freedom from protein.

Colloidal precipitating agents have come into extensive use in biological chemistry and the application of these methods has overcome one of the chief difficulties in the determination of sugar in blood.[4] For their use in nitrogen precipitations the chief inconvenience is that the blood or serum must be much diluted, which gives too large a filtrate volume for convenience in routine work. When, however, nitrogen partitions are being carried out with the filtrate, the volume is not an obstacle. It is possible that of some nitrogen-containing substances found in blood a small part may be adsorbed in the coagulum. There is no clear-cut evidence for this, however, as the values for total nitrogen check well with other methods. In practice two substances may be employed as precipitating agents, kaolin and dialyzed iron (Ferrum oxidum dialyzatum). Kaolin may be used with serum, but does not work well when hemoglobin is present; with whole blood it is necessary to resort to iron. The blood is diluted, one part to twenty of water; of a 10 per cent. solution of the iron there is added 2 to 3 c.c. for each c.c. of blood used, shaking constantly. The flask is allowed to stand half an hour then some electrolyte added—a gram of sodium sulphate. The volume is made up to mark, let stand an hour and then filtered.

For routine work a convenient method is the use of uranium acetate. The serum is diluted three times with water and an equal volume of 1.5 per cent. uranium acetate added. A water clear filtrate results, free of protein. In the dilutions employed uranium precipitates none of the crystalloids of blood.[5]

The method of determining the total nitrogen generally employed is the Kjeldahl. For urea Benedict's procedure, or Van Slyke's urease method, is most convenient, and gives dependable results. The various

4. Rona and Michaelis: Biochem. Ztschr., 1908, vii, 329; viii, 121.
5. Aszacki: Zentralbl. f. inn. Med., 1912, xlvii, 1166.

hypobromite methods still used by some French clinicians are wholly untrustworthy.

The non-protein nitrogen, or as it has been variously called, "filtrate" nitrogen, "rest" nitrogen, is a sum of several fractions, not all of which have been identified. As some of these substances may be increased in specific diseases without materially affecting the total (since nitrogen is but one element in the substance), our theme may be considered from several points of view. For example, in relation to uric acid, an uricemia has been noted quite uniformly in gout, pneumonia, leukemia, and with some cases of nephritis; and with all of these there is not necessarily a significant rise of the total non-protein nitrogen above normal, although this may occur in some instances. Likewise considerable rise in the values for ammonia-nitrogen may occur without appreciably influencing the total. From this it is evident that investigations may not be confined to total nitrogen but must include a consideration of the integral factors composing the non-protein nitrogen of blood.

With normal men the total non-protein nitrogen appears never to rise much over 40 mg.; or to fall much below 20 mg. per one hundred c.c. of blood, and the largest part of this is urea-nitrogen which fluctuates enormously. The extremes in my determinations are 92 and 46 per cent. urea-nitrogen for men apparently normal; more commonly 60 to 70 per cent. In order to avoid the influence of ingested proteins it has been my practice to take the blood in the morning between 10 and 11 o'clock, the hospital breakfasts containing but little protein.

Appreciable increases in the non-protein nitrogen in the blood have been observed in a number of different diseases besides nephritis. The rise accompanying cholera can possibly be explained in part as a result of the oligemia which is conspicuous in both the Asiatic and nostras types, and with the former there is frequently a complicating nephritis demonstrable at autopsy. With pneumonia an increase in the non-protein nitrogen has been in my experience exceptional and associated only with marked evidence of circulatory disturbance. Herter observed an increased urea content in the blood of some patients with pneumonia and suggested that all such cases be comprised under the term "uremia." The conspicuous cardiac disturbance with those cases of pneumonia in which the filtrate nitrogen was found abnormal, seemed possibly to explain the accumulation on a circulatory basis and led me to an investigation of some cases of pure decompensation due to valvular disease. In selecting cases for this series only young individuals were utilized. Of the series here reported the ages ran between 18 and 26 years. It was attempted further to exclude

those cases in which an antedating disease such as scarlet fever would lend an etiological basis for a persistent renal change. The cases in this series all presented extreme degrees of decompensation when they were admitted to the hospital; cyanosis, dyspnea, edema, hydrothorax, congested and often pulsating livers, oliguria. An initial albuminuria usually subsided or vanished when the cardiac condition improved. It has usually been stated that broken compensation, unassociated with renal change, does not occasion an increase in the non-protein nitrogen of the blood, and with the lesser degrees of circulatory embarrassment the facts do not run counter to theory. But with the severest manifestations of this disorder it would be difficult to understand how those conditions which bring about in some cases almost or complete anuria should not also lead to a heaping up in the blood-stream of the materials excreted normally through the kidney. There is also with these cases an increased katabolism coincident with the fever which is almost invariably present.

TABLE 1.—NON-PROTEIN NITROGEN FINDINGS IN CIRCULATORY DISTURBANCES NOT NEPHRITIC

| Diagnosis | Non-Protein N. |
|---|---|
| 1. Cardiac, valvular | 40 |
| 2. Cardiac, mitral stenosis | 59† |
| 3. Cardiac, aortic insufficiency | 61† |
| 4. Cardiac, aortic insufficiency | 47 |
| 5. Cardiac, mitral stenosis; cerebral embolism | 53 |
| 6. Cardiac, mitral and tricuspid | 73† and 85 |
| 7. Cardiac, mitral and tricuspid | 90† |
| 8. Cardiac, aortic | 43 |

† Necropsy.

The cases reported in Table 1 suggest, at least, that an increase in the filtrate nitrogen can be a result of purely circulatory disturbances. In but four of these cases was the absence of a nephritis confirmed by necropsy. The kidneys in these instances presented the typical picture of cyanotic induration. The cell infiltration was relatively slight and the epithelial change just apparent. The average non-protein nitrogen in the cases reported is 61 mg. per hundred c.c. of blood with 40 mg. and 90 mg. as the extremes.

If these results can be confirmed by other students of these diseases the fact involved is important as indicating the part played by the circulation in a picture already recognized as composite. It also has relations to diagnosis.

The significance of the non-protein nitrogen in renal diseases may be considered from a number of aspects; concerning its bearing on diagnosis and prognosis much has already been written and too often not in a judicial tone. The relation to anatomical and physiological conditions has been less considered primarily due to the fact that

nephritis in man involves all the renal structures to some extent and the physiological questions are complicated by a subtle perversion of the whole metabolism.

The following cases are some of those observed during the last two years and are selected on the basis of completeness of the clinical data secured during the period of observation in hospital. In most instances a number of blood determinations have been made in each case and when these vary, one from another, to a significant degree they are recorded.

TABLE 2.—NON-PROTEIN NITROGEN IN NEPHRITIS WITH AND WITHOUT UREMIA

| Case No. | Non-Protein N. | B. P. | Remarks |
|---|---|---|---|
| **PARENCHYMATOUS NEPHRITIS WITHOUT UREMIA** | | | |
| 19 | 61 | 160-190 | Post-pneumonic. |
| 101 | 62 | 100-130 | Edema, anemia. |
| | 74 | | |
| | 71 | | |
| | 70 | | |
| 104 | 61 | 140 | Edema. |
| | 43 | | |
| 107 | 70 | 120-140 | |
| **WITH UREMIA** | | | |
| 5 | 39 | ..... | Oliguria, edema, convulsions. |
| 131 | 115 | ..... | Convulsions; at autopsy, large white kidney. |
| **CHRONIC INTERSTITIAL NEPHRITIS WITHOUT UREMIA** | | | |
| 1 | 38 | 150-70 | Cardiac symptoms. |
| | 42 | | |
| 2 | 45 | 150-160 | Lassitude, gout. |
| 3 | 74 | 160-180 | Edema. |
| | 70 | | |
| 6 | 64 | 180-220 | Headache; died in two months. |
| 7 | 14 | 200 | Headache, edema; died in three weeks. |
| | 22 | | |
| 8 | 47 | 180 | Dyspnea, nausea. |
| 52 | 132 | 200-150 | Dyspnea, edema; died after five months. |
| (one month later) | 45 | | |
| 57 | 48 | 170-220 | Headache, dyspnea; died. |
| 58 | 120 | 200 | Edema. |

Of the first group only exceptional cases are here recorded. These cases of parenchymatous nephritis indicate that the non-protein nitrogen in this type of renal lesion is not necessarily low; Case 101 was of particular interest. The patient was kept on a weighed and analyzed diet (7 to 8 gm. of nitrogen per day) for over a month. At the time of his first determination (62 mg.) he was in bed and there was massive edema. There was a rapid loss of water and a subsidence of the edema which was accompanied by a rise in the non-protein nitrogen notwithstanding a large negative nitrogen balance.

The special interest that resides in cases of this character is the relation the "rest" nitrogen holds to water accumulations in the body. It has been suggested in explaining the relatively slight rises of the non-protein nitrogen with parenchymatous nephritis that the water retention effects a dilution and hence masking of the true state of affairs.

Case 131 was thought on account of the high non-protein nitrogen to be one of chronic interstitial nephritis with an acute exacerbation to explain the urine picture. The necropsy revealed a typical large white kidney.

As a rule, my results with parenchymatous nephritis agree with those of other investigators. The lowest figure is 32 mg. and the highest 115 mg. (uremia, Case 131) with an average of 63 mg. When uremia occurs, I have the impression that the "rest" nitrogen is often 150 mg. or over, since a number of cases of uremia in which high values were found were apparently due to parenchymatous nephritis; but in the absence of pathological examination proof is wanting. Strauss found an average of from 62 to 63 mg. in four cases of this type of renal lesion, with uremia and 80 mg. as a maximum. With the transitional types of nephritis there is at once an increase of values up to 120 mg. as an average in cases with uremic symptoms. Estimations of this character conducted with cases of nephritis in which there is water retention would be most instructive if they were combined with estimations of the whole blood volume. The significant observation of Strauss that edema fluids contain practically the same amounts of non-protein nitrogen as the blood has been adequately confirmed, and this fact offers an inviting explanation for the appearance of uremic symptoms with some cases following on the rapid loss of water (diuresis or diaphoresis). Case 101 illustrates the increased concentration that at times may occur even when there is a hyperazoturia, as the metabolism study indicated in this instance.

When we turn to the various types of contracted kidney the non-protein nitrogen tends to assume somewhat higher values, with notable exceptions, however, even in the uremic states. The average values for my first series of seventy-two cases was 87 mg., including cases of uremia; and this average has been reduced slightly, 84 mg., in the total determinations made to date in 130 cases. With cases free of uremic manifestations Strauss' average was 82 mg., which jumped to 130 mg. when there were uremic complications. Maximal determinations are recorded over 300 mg. without uremia (Folin) and below 50 mg. with uremic symptoms (Ascoli, Farr and Austin, Foster). An especial interest is attached to low values in interpreting the usefulness of these determinations as a diagnostic measure; the import of a high non-

protein nitrogen has been sufficiently explained in the literature, often ignoring or failing to recognize as uremia cases in which low values obtain. That severe grades of chronic nephritis do occur without appreciable rises of the "rest" nitrogen and that such patients may even develop a fatal uremia the accompanying tables attest. That these cases may not be the rule is granted, but I do not think they are as exceptional as they are commonly held to be. With frank uremic manifestations we encounter many cases in which the non-protein

TABLE 3.—Chronic Nephritis with Uremia

| Case | Nitrogen mg. | Blood-Pressure | Type | Termination |
|---|---|---|---|---|
| 1 | 68 | 190 + | Asthenic | Improved |
| 2 | 56 | 160 + | Asthenic | Died |
| 5 | 110 | 190 + | Conv. | Died |
| 7 | 62 | 150 | Conv. | Died, 1 mo. |
| 8 | 52 | 170 | Asthenic | Died, 6 mo. |
| 10 | 47 | 180 | Asthenic | Died |
| 14 | 72 | 220 | Coma | Died |
| 15 | 41 | 170 | Asthenic | Died |
| 16 | 198 | 250 + | Conv. | Died |
| 18 | 178 | 200 + | Conv. | Died |
| 22 | 65 | 160 + | Asthenic | Died |
| 23 | 104 | 160 | Asthenic | Died |
| 24 | 86 | 200 + | Conv. | Died |
| 26 | 40 | 280 | Asthenic | Died |
| 27 | 134 | 200 | Conv. | Died |
| .. | 142 | | | |
| 28 | 44 | 160 | Asthenic | Died |
| 29 | 41 | 210 | Asthenic | Died |
| | 70 | | | |
| 30 | 177 | 180 | Asthenic | Died |
| 31 | 21 | 200 | Asthenic | Improved |
| 32 | 136 | 210 | Conv. | Died |
| 33 | 100 | 160 | Asthenic | Improved |
| .. | 116 | | | |
| .. | 96 | | | |
| 34 | 239 | 190 | Conv. | Died |
| 36 | 61 | 100- 80 | Asthenic | Died |
| 43 | 98 | 120-130 | Stupor | Died |
| 47 | 75 | 170-200 | Conv. | Died |
| 60 | 46 | 200-250 | Stupor | Died, 1 wk. |

nitrogen is not higher than is observed with cases of latent nephritis quite free of renal symptoms. This becomes at once evident if one examines the blood of patients in the surgical wards who regard themselves as in perfect health except for the fracture or hernia which brought them to the hospital.

It is difficult to attempt to correlate the blood findings with any particular manifestations of the disease because of the variability of the latter, and this is particularly true of the blood-pressure, which

changes considerably from hour to hour. I have selected a fairly representative figure for systolic pressure from the day on which the blood was taken, or have used the low level and marked with a plus sign (Table 4).

A rough average struck by means of the data in Table 4 supports the current view at least in so far as it shows that the more pronounced degrees of nitrogen retention are prone to be associated with increased arterial systolic pressure.

TABLE 4.—VARIATIONS IN SYSTOLIC PRESSURE IN CASE WITH AND WITHOUT UREMIA

| Blood-Pressure, Mm. Hg. | Non-Protein Nitrogen, Mg. Per Cent. |
|---|---|
| WITHOUT UREMIA | |
| 150-170 | 42 |
| 170-190 | 63 |
| 190-210 | 67 |
| WITH UREMIA | |
| 150-170 | 73 |
| 170-190 | 78 |
| 190-210 | 120 |
| 210-250 | 108 |

It is of some moment to consider the types of uremic manifestations in association with the non-protein nitrogen. The symptoms of uremia are so various in their combinations that all the notable factors cannot be brought into this relation. Disregarding subordinate considerations, uremia is sharply divided into two classes by the presence or absence of convulsive seizures. When convulsions are wanting, the notable element in the symptom-complex sooner or later is asthenia, physical and mental, which grades off into stupor and coma. In Table 3 there are nine cases of the epileptiform type of uremia and seventeen of the asthenic—about the relation they bear to each other in incidence. For the convulsive cases the non-protein nitrogen averages 135 mg.; the extremes being 62 mg. and 239 mg. For the asthenic types the average is 67 mg., with 21 mg. and 177 mg. as the low and high values. There appears, then, to be some relation which the non-protein nitrogen holds to the character of the uremic manifestation; the higher values are observed more commonly in epileptiform uremia.

Of the uremic patients whose cases are here reported only three lived longer than six months; hence it is evident that we can draw no conclusions from these data as to the prognostic significance of the "rest" nitrogen in uremic states. In the absence of uremic symptoms, the non-protein nitrogen has, when the values are high, a definite prognostic significance.[6] With over 100 mg. the outlook is extremely grave.

6. Foster, Nellis B.: Pathological Deviations in the Chemistry of Uremic Blood, THE ARCHIVES INT. MED., November, 1912, p. 414.

But this fact applies only to a fraction of the cases examined and it cannot be overlooked that values which are but slightly above normal fail to exclude a not remote fatal issue. Table 3 supports this statement in regard to uremia.

Thus far the non-protein nitrogen has been considered as a whole. Its composition is of interest both clinically and pathologically. In a former paper[6] data were given indicating the fluctuation of the urea fraction of the total nitrogen as between 35 and 80 per cent., and also some determinations of the purins and nitrogenized lipoids. These investigations have been broadened to include all of those nitrogen-containing substances known to occur in urine, since it seemed highly desirable to have better knowledge concerning the catabolic products influenced by diminished renal function, and also at the same time, any other substances that make up the total nitrogen with those cases of uremia in which the urea-nitrogen forms but half or less of the total nitrogen.

For the estimation of ammonia in blood the customary aeration method has been employed. As the amounts of ammonia are small, it is necessary to use hundredth-normal acid and alkali. Excellent controls are secured with iodo-eosin in ether as an indicator. The amount of ammonia-nitrogen in normal blood seldom rises above 0.5 mg. per hundred c.c. by this method. Winterberg[7] found 0.6 mg. and 1.3 mg. as the limits of normal, depending on the influence of taking food. Folin's[8] results average 0.1 mg.

There is a decided increase in the ammonia of blood with many diseased states, the highest values having been noted with diabetic coma. Winterberg also observed 2 mg. or more with infectious diseases. With the bloods of uremic patients my results may best be summarized; 28 cases were examined. In 11 instances the ammonia-nitrogen was below 0.5 mg.; that is, normal. With 12 the values were between 0.7 and 1.6 mg.; and with 3 between 1.8 and 2.2 mg. per hundred c.c. of blood. The highest values were found with cases of the convulsive type of uremia when the total nitrogen was high. Winterberg found slightly over 2 mg. of ammonia nitrogen with two cases of uremia, while Folin's figures range from 0.6 to 1.0 mg. for uremic bloods. The net result of these estimations indicates that ammonia accumulates in the blood in something over half of the cases of nephritis with uremia, and that exceptionally this increase may be considerable; but it tends to bear a normal relation to the total non-protein nitrogen.

7. Winterberg: Ztschr. f. klin. Med., xxxv.
8. Folin: Jour. Biol. Chem., 1914, xvii, 488.

The earlier estimations of the uric acid in blood are of doubtful accuracy on account of the method employed, yet the opinion has been long held, and is borne out by the recent work of Folin and others, that with renal disease there is uric acid retention. For the purpose of my study it seemed more advisable to determine the total purin content of the blood by a double precipitation with the Kruger-Schmidt method. The purin-nitrogen of normal blood varies from 3 mg. to 6 mg. per hundred c.c. of blood. With nephritis the average is higher than normal, although the increase with the parenchymatous types is much less apt to occur and is of less pronounced degree than with contracted kidney. With uremic patients the purin-nitrogen may reach 16 mg. in exceptional instances; the average for ten cases was 12 mg. in my series. No relation whatever can be detected between the total non-protein nitrogen and the purin nitrogen. The highest values for the latter may accompany only moderate increases of the total nitrogen, and in one case with a high total, 311 mg., the purin nitrogen was but 7 mg. Folin's[9] estimations of uric acid in relation to the total nitrogen show the same absence of concordance.

Estimations of creatin and creatinin were made in but four instances, as sufficient blood was not available with other cases. The proteins were separated out by the adsorption method of Rona and the filtrate concentrated. After hydrolysis with acid the readings were made using Shaffer's modification of the Folin method. The four cases studied were all of the convulsive type. The estimations in these four cases were 28 mg., 27 mg., 32 mg. and 46 mg., respectively. There is then so far as this evidence goes a considerable retention of either creatin, creatinin, or both as result of the renal lesion.[10]

It remains to consider the amino-acids, four estimations of which were done by the Van Slyke method.[11] The four cases were selected so as to represent various degrees of accumulation of non-protein nitrogen from 188 mg. down to normal. The amino-acid content was normal in all, 4 to 6 mg.

The history of the theories of causation of uremia is an epitome of the supposed toxic action of the various fractions which compose the non-protein nitrogen of blood. Each of these substances is in a certain degree toxic but not in the concentrations found in body fluids. While it is conceivable that certain cells, as, for example, nerve cells, may under some circumstances, exhibit, as Strauss has recently suggested, an increased irritability to these agents, yet such an idea is purely conjectural. That none of the substances with which we are

9. Folin: Jour. Biol. Chem., 1914, xvii, 489.
10. Myers and Fine: Proc. Soc. Exper. Biol. and Med., 1914, xi, 132.
11. Due to the courtesy of Dr. Donald Van Slyke.

familiar is to be held accountable for uremia is attested by excessive accumulations of these substances in certain morbid states without characteristic symptoms. Herter analyzed forty-one cases of anuria (not nephritis) recorded in the literature; with seven the anuria persisted for fourteen days or longer; and with these no symptom which could be regarded as uremic appeared before the eleventh day. It is well known that cases of poisoning with bichlorid of mercury present very few symptoms other than those due directly to the poison; yet the blood in such cases has yielded some of the highest values for "rest" nitrogen recorded. Whatever may be the significance of the known fractions composing the non-protein nitrogen, clinically or pathologically, it seems highly improbable that they are directly related as a cause to uremic manifestations. With some cases of uremia the sum of these known fractions practically equals the total nitrogen; while with other cases there is an appreciable unknown, undetermined fraction. Studies on this point are now in progress.

Concerning the location of renal lesions which induce faulty nitrogen elimination we have no conclusive evidence. With clinical nephritis of even the least complex types there is enough involvement of all the renal structures to make conclusions as to function hazardous. Experimentally induced nephritis offers some interesting data which it is difficult in many instances to harmonize with the clinical facts. There is an increase in the non-protein nitrogen with nephritis caused by poisoning with uranium, chromium and tartaric acid. All of these produce[12] lesions which are almost wholly tubular, any glomerular implication being inconstant and of slight degree. Arsenic involves both glomeruli and tubules, the lesions of the latter are earlier and more transitory than in the glomerulus; and according to Karsner it is during the period when the tubules are suffering, up to seventy-two hours after administering the poison, that the non-protein nitrogen shows its largest values. The highest values were also noted in association with the greatest tubular damage. The nearest approach to a pure glomerular nephritis that can be experimentally induced is by means of diphtheria toxin, but there is here also cloudy swelling and focal necrosis in the tubules. There is with this form of nephritis also considerable increase in the non-protein nitrogen of the blood.

The trend of evidence points to tubular rather than glomerular involvement as the important factor in nitrogen retention, since the phenomenon is clearly manifested with experimental renal lesions that are almost, if not entirely, epithelial. It is then of considerable interest that the nearest approach to a purely tubular nephritis clinically is not

12. Folin, Karsner and Denis: Jour. Exper. Med., 1912, xvi, 789. Karsner and Denis: Jour. Exper. Med., 1914, xix, 263.

marked by the highest degree of nitrogen retention. (The possible deception due to edema has been already noted.) There arises then the question of degrees of structural damage in relation to function. With the contracted kidney there is so much destruction of all renal elements that it becomes quite impossible to assert that one structure is more damaged than another.

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# THE EFFECT OF ANESTHESIA AND OPERATION ON THE KIDNEY FUNCTION, AS SHOWN BY THE PHENOLSULPHONEPHTHALEIN TEST *

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The question of the effect of anesthetics on the kidney has long been one of interest, and it seemed to us that an investigation of this by means of the phenolsulphonephthalein test of Rowntree and Geraghty would be of considerable interest. No extensive investigation of the subject in this manner has come to our attention, though a brief report is given by Sanford[1] who did the test in a series of cases of ether and gas-oxygen anesthesia, doing it first twenty-four hours before operation and again within a few hours afterward. He says:

> The cases of gas anesthesia gave almost the same output of phthalein after anesthesia as before, showing apparently no effect on the kidney from the anesthetic. The ether cases have been rather contradictory. In short anesthetics the output of the drug was almost doubled after ether, and in long operations it was generally diminished. It seems probable that a short ether anesthetic is a renal stimulant, while a longer one is a depressant. So many factors enter into a renal functional test done within a few hours after an operation, that this can only be a conjecture.

Also in a recent article Block[2] reports about twenty cases tested with phenolsulphonephthalein before and after operation, and says that although the series is too small to serve as a basis for definite conclusions, it seems to indicate, among other things, that the effect of anesthesia on healthy kidneys is practically nil so far as reducing the excretory power goes; the duration of anesthesia in any given case does not of itself signify the amount of injury which the kidneys will sustain; the excretion of phenolsulphonephthalein after, as compared with before operation, is not affected by the administration of proctoclysis, the position of the patient on the table or the character of the operation; and that in about one-third of the cases there is increased excretion after operation.

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* Submitted for publication June 22, 1914.

1. Sanford: A Clinical Study of the Elimination of Phenolsulphonephthalein by the Kidneys, with a Report of 150 Cases, Cleveland Med. Jour., 1912, xi, 763.

2. Block, Frank Benton: The Effect of Operation on Kidney Excretion as Indicated by the Phenolsulphonephthalein Test, Jour. Am. Med. Assn., April 25, 1914, p. 1309.

A great many articles have appeared on the effect of anesthetics on the kidney as shown by the presence of albumin, blood, pus and renal elements, and to these brief reference will be made. Bovée[3] found that during the first fifteen minutes of ether anesthesia there was an increase in the output of urine, followed by a decrease up to forty-five minutes, after which it increased again. The diminution in urea output was even greater in proportion than that of the urine. He says that ether, carefully and skilfully given, has little effect on the production of albumin and casts, inducing them in some cases and stopping them in others. He states that in the Trendelenburg position there is even greater interference with urinary excretion, due, he thinks, partly to retraction in the renal pelvis and partly to a certain degree of arrest of renal function directly due to the position. Hewitt[4] has collected interesting figures from various authors as follows:

Weir..... 34 ethers, followed by 9 cases of albuminuria
Barenfeld.....150 ethers, followed by 2 cases of albuminuria
Butler .....500 ethers, followed by 1 case of albuminuria
Kute.... .....600 ethers, followed by 1 case of albuminuria

Hewitt, summing up the question, says:

With regard to the frequency of renal complications after ether it is difficult to speak with certainty, owing to the fact that statements made by those observers who have specially studied this point are very conflicting. The author has not, to his knowledge, met with any cases in which such complications have occurred, but it is only fair to say that anesthetists' opportunities for ascertaining the condition of patients after anesthesia are very limited.

Buxton and Levy (quoted by Hewitt[4]) are "unable to satisfy themselves that ether, when properly administered, exerts any unfavorable influence on the kidneys"; they say that such ill effects occur chiefly, if not wholly, in cases in which the ether has been unduly and unnecessarily pushed. Kemp (quoted by Hewitt[4]) says that ether causes a special contraction of the arterioles of the kidney, with decreased secretion of urine. He believes that it gives rise to a damaged state of the secreting cells and that albumin appears early in the urine secreted.

Regarding the purely psychic effect of the ordeal of preparing for operation and taking the anesthetic, the following quotation from Crile[5] is of interest:

We now know that fear alone may cause physical damage, not alone to the brain, but to other organs. It causes an increase in adrenalin, in the output of glycogen, and in rabbits repeatedly frightened, albumin and casts are sometimes produced.

3. Bovée: Renal Excretion During the Administration of Chloroform and Ether in Gynecological Surgical Operations, Tr. New York Obst. Soc., Am. Jour. Obst., 1909, lix, 1004.
4. Hewitt: Anesthesia, Ed. 4, 1912, p. 383.
5. Crile: Nitrous Oxide Anaesthesia, with Note on Anoci-Association, a New Principle in Operative Surgery, Surg., Gynec. and Obst., 1911, xiii, 170.

Von Brunn[6] says that in deep anesthesia the urine is decreased in amount, though occasionally it may show increase, and that for two or three days there is an increase in the specific gravity. The loss of function of the renal parenchyma apparently corresponds to the length of the anesthesia (compare our cases below). He says that ether may, if given several times within twenty-four hours, cause fatty degeneration and necrosis of tissues in animals. He has collected figures from various authors, taking into consideration the occurrence of albumin before as well as after operation, and these will be stated in part:

| | Albumin Before | Albumin After |
|---|---|---|
| Roux— | | |
| 119 cases ether | 4 | 4 |
| Butter— | | |
| 500 cases ether | 0 | 1 |
| Wunderlich— | | |
| 72 cases ether | 0 | 6 |
| 52 cases chloroform | 0 | 6 |
| 1 case chlor-ether | 0 | 1 |
| Eisendrath— | | |
| 60 cases ether | 25.0 per cent. showed albumin afterward | |
| 70 cases chloroform | 32.0 per cent. showed albumin afterward | |
| Deaver and Frese— | | |
| 63 cases ether | 47.6 per cent. showed albumin afterward | |

Von Brunn says:

> It must be said that ether may cause trouble with the kidney, which may manifest itself by irregularities of excretion, decreased nitrogen excretion, albuminuria and cylindruria. These, however, are only of a fleeting nature, and occur, at least so far as albumin and casts go, in only a small percentage of the cases. A preexisting albuminuria or cylindruria may be increased by ether narcoses. Compared with chloroform, ether is less dangerous.

In nephritis, general narcosis should be avoided if possible — if it cannot be avoided, ether is better than chloroform.

Popoff[7] reported 140 cases of patients examined before and after ether. Of these 140 cases, 14 showed albumin before operation and 28 afterward — in other words, 10 per cent. had albuminuria as a result of operation under ether. Konwer (quoted by Popoff[7]) reports 100 cases of which 5 showed postoperative albuminuria. Popoff concluded that ether may cause transient albuminuria, that it may cause or increase casts, but that nevertheless the general condition of a patient is never made worse by it. Buxton,[8] as a result of his investigations, states that:

> In most cases the quantity of the urinary water was reduced, but probably not more so than could be accounted for by the abstinence from food and liquids. The solids remained practically unaffected. When ether is given in

6. Brunn, M. V.: Die Allgemeinnarkose, Tübingen, 1913, p. 171.
7. Popoff, Wassil: Contributions à l'étude de l'albuminurie après l'éthérisation, Thèse Inaugurale, Genève, 1896.
8. Buxton: Anaesthetics, Philadelphia, 1907, pp. 47, 120.

excessive quantity, ischemia of the kidneys is produced and albuminuria results. If, however, only so much is inhaled as is needed for complete anesthesia, no deleterious effects arise, and the renal parenchyma is not injured nor does albuminuria result. The experimental work was supplemented by the study of clinical cases and these bore out our experimental deductions.

Later he says:

In renal disease, when extensive, ether is said to induce suppression of urine, so that if given at all in these cases it should be used with caution.

Blake[9] gives figures collected by other authors concerning death from anesthesia, and those are interesting in this connection:

| | | |
|---|---|---|
| Gurlt— | | |
| 166,812 | chloroform cases, 63 deaths | 1 in 2,647 |
| 26,320 | ether cases, 2 deaths | 1 in 13,160 |
| 8,014 | ether and chloroform, 1 death | 1 in 8,014 |
| Zeigler, | 600 ether cases | No deaths |
| Tschmarke, | 500 ether cases | No deaths |
| Körte, | 600 ether cases | No deaths |
| Vogel, | 1,200 ether cases | Two deaths or serious complications |
| Ollier, | 40,000 ether cases | No deaths |
| Chalot, | 730 ether cases | No deaths |
| Tillier, | 6,500 ether cases | No deaths |
| Poucet, | 15,000 ether cases | No deaths |
| Juillard— | | |
| 314,738 | ether cases, 21 deaths | 1 in 14,987 |
| 524,507 | chloroform cases, 161 deaths | 1 in 3,258 |

Blake examined personally 50 cases before and after ether; of these, 17 showed albumin before, and in 11 of these 17, the albumin was increased. Of the 33 with no albumin before operation, 25 showed it afterward. These figures are much higher than those of most other observers. Blake believes that the albuminuria is due to a passing irritation of the kidney and not to a true nephritis.

J. Wyllis Hassler,[10] speaking in regard to the urine after intravenous anesthesia, says:

The urinary analysis made before the operation and for three or four days afterward showed no marked difference in the specimens. The total amount was increased and the specific gravity lowered in the first twenty-four hours; often the specific gravity and solid content of the urine remained unchanged. In no case, even after the employment of a 7.5 ether solution, did blood, albumin or casts appear in the urine, though German observers did report occasional cases of transient hemoglobinuria after the use of the stronger ether mixtures.

There are countless articles in the literature dealing with one or another of the dangers of anesthetics and how to avoid these dangers, but as the great majority have no direct bearing on the subject in hand, no further reference will be made to them.

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9. Blake, J. B.: An Examination of Some Recent Statistics in Regard to Ether, and a Consideration of Some Present Methods of Its Administration, Boston Med. and Surg. Jour., 1895, cxxxii, 559.

10. Hassler: Intravenous Anaesthesia, Ann. Surg., 1913, lviii, 900.

## AUTHORS' WORK

Our work was done on the ward cases at the Massachusetts General Hospital, the patients being taken without regard to age, general condition, or the specific nature of their ailment. A desire for as much accuracy as possible made it seem wise to avoid, to a certain extent, such nephrectomies and nephrotomies as would have, after operation, an excess of blood in the urine, and the prostatectomies, in which the constant irrigation after operation, plus the blood, made careful work well-nigh impossible. Also it was found impossible to use stricture cases on account of the difficulty of getting the urine before operation. After early experiences in the work, attempts to carry through the test on female patients were given up on account of the unavoidable difficulty encountered in getting them catheterized at the proper time after operation. This was not due in the least to any unwillingness on the part of the attendants, but to other duties which continually presented themselves and which were clearly too important to neglect. The collection of the urine at the end of the two-hour period was done personally, as it was early found that this was the only way to be sure of having it done completely and at the proper time.

The great majority of the patients were tested on the day of their entrance into the hospital — only a small number being tested after they had been in the wards one or more days. Whether the fact that most of the patients were more or less frightened over their new surroundings and anticipations of future operation could have any reflex inhibitory effect on their visceral functions and more particularly on their kidney function, as shown by phenolsulphonephthalein, would be in most cases, difficult to state — though such a possibility does not seem beyond the realms of reason (see Crile[5] quoted above).

Each patient was directed to empty his bladder and was then given 1 c.c. of phenolsulphonephthalein into the muscles of the thigh. Injections into the lumbar muscles were given up early on account of the difficulty encountered in attempting to give the injection in the same place after the operation. The patients were questioned as to the frequency of micturition, nocturia, etc., and those in whom was no history suggesting retention and residual urine emptied their bladders by voiding at the end of two hours from the time of injection. In only a very few cases did catheterization seem necessary at this time, but as will later be pointed out, certain discrepancies in some of the results are most probably due to the fact that the patient really did not completely empty his bladder, though he seemed to at the time. The drug was given with a 3 c.c. Paris glass syringe, the plunger being drawn up to the mark 1.1 c.c., then driven in to the mark 0.1 c.c., thus obviating any error from injecting bubbles of air, which would otherwise occasionally happen.

The second injections were given at various times — in some cases just at the beginning of the operation, and in others at different periods after the end of the operation — some being a day or more later. At the end of the two-hour period, if the patient was perfectly conscious and could void without difficulty, he was allowed to do so, but in the majority of cases the catheter was necessary. All the ordinary aseptic precautions were taken in catheterizing, and to the best of our knowledge in no case was the bladder or urethra infected. Also, no patient was infected from the injecting needle, though occasionally a patient would complain for a day or two of a stiff, sore thigh. In catheterizing patients who were still under ether, or only partly out of its influence, a large number were found to have a profuse secretion of mucus from the urethra, in many cases almost enough to lubricate the catheter. This secretion was observed in almost every case in which the foreskin covered the glans; in the other cases it undoubtedly was present, but was wiped away as the patient moved about in bed. This seems to prove that ether causes the secretion of mucus from the urethral mucous membrane, and not improbably from all the mucous membranes of the body, though usually its effect is apparent only in the upper air passages and membranes which come in direct contact with the ether vapor.

The attempt was always made to make the collection at the end of exactly two hours, but an error of a very few minutes — practically never over five minutes — was frequently unavoidable, owing to various factors, but it did not seem to us that an error so small as that would make any material difference in the results.

The two-hour specimen was measured for the amount, and the reaction noted; it was then made alkaline with 33 per cent. potassium hydroxid and diluted to 1 liter and filtered. It was then compared in the Duboscq colorimeter with a standard solution of 3.0 mg. phenolsulphonephthalein to the liter of water, and made slightly alkaline. In practically none of the cases was the urinary pigment abundant enough to cause doubt as to the accuracy of the reading. In a few cases in which there was considerable blood we found one fairly satisfactory method was to let the specimen stand over night, at the end of which time most of the blood would be settled at the bottom of the flask, the overlying urine being fairly clear and only slightly stained with blood pigment. Part of this clear urine was decanted off and examined, and then the total phenolsulphonephthalein content estimated by measuring the two portions and figuring out how much would be contained in the rest of the urine in the flask. This seems to be a reasonably efficient method. In one case in which there was an excess

of bile in all specimens, the urine obtained before operation was used as a standard with which to compare that obtained afterward, and in this way a fairly accurate comparison could be made. For a long time artificial light was not used, but toward the end of the work it was discovered that the readings made by electric light were always the same as those made by daylight, and thereafter there was no hesitation about doing the readings by electricity. (The feasibility of this use of artificial light is agreed to by Dr. Palmer of the chemical laboratory at the hospital.)

Coming to the actual results of the work, it is of interest first to consider the average output of the patients just as they came to the hospital, and before operation. No account is taken in these figures of the patient's condition or that of his kidneys, though in the great majority of the cases the kidneys were normal. These figures also do not include a few cases, which will be referred to next, in which more than one test was done, but there was no operation. There were 422 such cases with an average output in two hours of 58.5 per cent. This figure is lower than that of Rowntree and Geraghty,[11] who state that the normal is from 60 to 80 per cent., but they measure from the time of appearance, and not from the time of injection, as in this case. On the other hand, this is higher than that of Keyes and Stevens,[12] who get an average normal output, in two hours from the time of appearance, of only 57.2 per cent. In this series the highest total for the two hours was 94.0 per cent., and the lowest 00.0 per cent. The latter was in the case of a much-emaciated and cachectic woman with inoperable carcinoma of the cervix uteri. She failed to excrete any of the drug in two hours after a subcutaneous injection, and the following day failed to excrete any in one hour following intramuscular injection. She subsequently went through an operation under gas and oxygen for cautery of the growth, and then was lost sight of. One urinalysis was as follows:

Pale, turbid, acid, 1.010; albumen, trace; sugar, 0; sediment, much pus.

and two days later:

Milky, acid, 1.014; albumin, very large trace; sugar, 0; sediment, much pus.

She survived the operation and left the hospital. She has undoubtedly died of cancer before now, if not from renal insufficiency.

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11. Rowntree and Geraghty: An Experimental and Clinical Study of the Functional Activity of the Kidneys by Means of Phenolsulphonephthalein, Jour. Pharmacol. and Exper. Therap., 1910, 1, 579.

12. Keyes and Stevens: A Clinical Study of Renal Function by Means of Phenolsulphonephthalein, Am. Jour. Urol., 1911, vii, 367.

Of the above-mentioned 422 cases, 409 were next classed into four groups according to ages, as follows:

Group 1, age, 1-19, inclusive
Group 2, age, 20-39, inclusive
Group 3, age, 40-59, inclusive
Group 4, age, 60-.., inclusive

Their output was then estimated according to these groups, and in spite of the fact that, as noted above, no account is taken of their condition, the results are interesting:

Group 1, age 1-19, 46 cases.
Lowest, 27.0 per cent.; highest, 90.9; average, 67.8 per cent.
Group 2, age 30-39, 173 cases.
Lowest, 20.0 per cent.; highest, 92.6 per cent.; average, 62.2 per cent.
Group 3, age 40-59, 154 cases.
Lowest, 00.0 per cent.; highest, 80.5 per cent.; average, 56.8 per cent.
Group 4, age 60, 36 cases.
Lowest, 5.0 per cent.; highest, 82.5 per cent.; average, 45.7 per cent.

To put it more concisely:

Age 1-19, average phenolsulphonephthalein, 67.8 per cent.
Age 20-39, average phenolsulphonephthalein, 62.2 per cent.
Age 40-59, average phenolsulphonephthalein, 56.8 per cent.
Age 60 .., average phenolsulphonephthalein, 45.7 per cent.

These figures illustrate rather strikingly what one might be led to expect, namely, that the kidney function, as demonstrated by the excretion of phenolsulphonephthalein, becomes progressively less as one grows older.

Before taking up variations in function following anesthesia, it is of interest to consider a few cases in which the functional test was performed more than once on the same patient, without operation. This will give an idea of how much fluctuation in the test one may expect under ordinary circumstances. A few cases taken from other authors will first be mentioned:

Rowntree and Geraghty.[13]

Intravenous injection, output in one hour:
Case 5, Test 1, 62.5 per cent.
Test 2, 62.5 per cent.
Test 3, 63.3 per cent.
Case 10, Test 1, 64.5 per cent.
Test 2, 72.7 per cent.
Case 12, Test 1, 62.5 per cent.
Test 2, 65.8 per cent.
Test 3, 65.5 per cent.
Case 14, Test 1, 71.5 per cent.
Test 2, 68.5 per cent.

13. Rowntree, L. G., and Geraghty, J. T.: The Phthalein Test, THE ARCHIVES INT. MED., 1912, ix, 284.

Lumbar injection, one-hour excretion:

Case 6, Test 1, 54.0 per cent.
Test 2, 54.3 per cent.
Case 10, Test 1, 40.2 per cent.
Test 2, 60.9 per cent.
Test 3, 60.2 per cent.
Test 4, 60.2 per cent.
Case 11, Test 1, 61.7 per cent.
Test 2, 64.1 per cent.
Test 3, 63.2 per cent.
Case 13, Test 1, 62.5 per cent.
Test 2, 62.5 per cent.

## Cases of Keyes and Stevens.

Comparison of excretion on different days:

Case 1.—Syphilis of spinal cord:
Test 1: Appear. time, 8 minutes, two-hour excretion, 71.2 per cent.
Test 2: Appear. time, 9 minutes, two-hour excretion, 56.0 per cent.
No change of condition clinically.

Case 2.—Hypospadias:
Test 1: Two-hour excretion, 56.0 per cent.
Test 2: Two-hour excretion, 49.4 per cent.
Test 3: Two-hour excretion, 61.8 per cent.
Test 4: Two-hour excretion, 56.0 per cent. (intravenous)

Case 3.—Gonorrheal epididymitis:
Test 1: Two-hour excretion, 54.6 per cent.
Test 2: Two-hour excretion, 53.+ per cent.

## Personal cases:

Case 1.—Man, aged 69, extensive rodent ulcer of face:
Test 1: April 30, 1913, two-hour excretion, 44.4 per cent.
Test 2: Aug. 12, 1913, two-hour excretion, 40.3 per cent.

Case 2 (No. 320).—Two tests done in the interval between an exploratory laparotomy and a later colostomy for cancer:
Test 1: July 2, 1913, two-hour excretion, 32.1 per cent.
Test 2: July 4, 1913, two-hour excretion, 33.3 per cent.

Case 3 (No. 349).—Man, aged 56; cancer of jaw:
Test 1: Two-hour excretion, 59.5 per cent.
Test 2: Two-hour excretion, 62.5 per cent.

Case 4.—Man; fracture of patella:
Test 1: Aug. 13, 1913, two-hour excretion, 71.4 per cent.
Test 2: Oct. 24, 1913, two-hour excretion, 67.5 per cent.

Case 5.—Man, aged 56; cancer of jaw; patient in poor condition:
Test 1: Sept. 17, 1913, two-hour excretion, 31.6 per cent.
Test 2: Sept. 20, 1913, two-hour excretion, 32.2 per cent.

Case 6.—Man; cancer of thyroid:
Test 1: October 7, two-hour excretion, 82.5 per cent.
This patient left the hospital and came back weak and emaciated, almost starved.
Test 2: November 6, two-hour excretion, 44.4 per cent.
In this case the change in the patient's condition might account for the decreased output.

Case 7.—Man, aged 31. Between the two following tests the patient was prepared for operation, given two enemas, and on the 21st starved up to the time of test:
Test 1: June 20, 1913, two-hour excretion, 54.9 per cent.
Test 2: June 21, 1913, two-hour excretion, 39.0 per cent.

Case 8.—Boy, aged 15; hernia. At the time of the first test this patient had bronchitis, because of which he was discharged without operation:
Test 1: March 18, two-hour excretion, 60.2 per cent.
Test 2: June 9, two-hour excretion, 79.3 per cent.

CASE 9.—A strong man, aged 28, in perfect health. The test was done at the same time each afternoon, but conditions had varied during the morning's work:

Test 1: June 10, two-hour excretion, 81.9 per cent.
Test 2: June 11, two-hour excretion, 71.4 per cent.
Test 3: June 12, two-hour excretion, 70.4 per cent.
Test 4: June 14, two-hour excretion, 76.9 per cent.
Test 5: June 15, two-hour excretion, 79.3 per cent.
Test 6: June 16, two-hour excretion, 70.4 per cent.
Test 7: June 17, two-hour excretion, 79.3 per cent.
Test 8: July 1, two-hour excretion, 71.4 per cent.*
Test 9: July 4, two-hour excretion, 82.5 per cent.
Test 10: July 6, two-hour excretion, 73.5 per cent.*

* Perspiring profusely.

These cases have been given in detail to show what fluctuation may be encountered, normally or abnormally. In certain of the foregoing there seems to be a definite reason for changes in the output, as in Case 6 of the authors. The figures of Rowntree and Geraghty are most consistent, though they have one variation of 20.7 and another of 8.20. Perhaps the most instructive figures are those of the last case, in which ten tests were done on a normal man, with results varying from 70.4 per cent. to 82.5 per cent. It seems that it would be a very difficult thing to name any limits which we could call normal for any given case.

### GENERAL AVERAGES BEFORE AND AFTER ETHER

These are the figures of 335 cases in which tests were performed on the entrance to the hospital and again after an operation under ether, with or without such accessories as nitrous oxid, anesthol (a mixture of ether, chloroform and ethyl chlorid), local anesthesia, etc. As will be noted later, not all the cases showed a decreased output of phenolsulphonephthalein, but they are included among these figures:

Number of cases, 335.
Average excretion, before operation, 59.3 per cent.
Average excretion, after operation, 48.1 per cent.

Difference ..... 11.2 per cent.
Decrease, 18.8 per cent. of average output before.

These are general figures, not taking into consideration the nature of the operation, the time of injection of the drug, or the condition of the patient.

### GENERAL AVERAGES BEFORE AND AFTER SPINAL ANESTHESIA

In this series there are not enough cases to prove of much value, but they are inserted here for what they are worth:

Number of cases, 16.
Excretion before operation, 49.7 per cent.
Excretion after operation, 41.4 per cent.

Difference ..... 8.3 per cent.
Decrease of 16.7 per cent. of average output before.

In spite of the few cases at hand, it is interesting that the decrease in phenolsulphonephthalein output is not so great as that after ether (18.8 per cent.).

LOCAL ANESTHESIA

The same may be said here as was noted in regard to the spinal anesthesia; there are only eight cases, which are interesting even if not of much value:

Number of cases, 8.
Excretion before operation, 50.8 per cent.
Excretion after operation, 44.7 per cent.

Difference ..... 6.1 per cent.
Decrease, 12.0 per cent. of output before.

Here again it will be noted that though few in number they show less decrease than the ether cases.

GAS AND OXYGEN

There were only nine such cases which we were able to test fully, and the results are given below, though they do not coincide with our theory as to what they should be. It would seem natural to expect the kidneys to show little or no effect after this anesthesia, whereas our few cases show even more decrease in output than after ether. A much larger number must be examined before any conclusions can be rightly drawn. These figures, therefore, are added only for what they may be worth:

Number of cases, 9.
Output before operation, 59.7 per cent.
Output after operation, 44.3 per cent.

Difference ..... 15.4 per cent.
Decrease, 25.7 per cent. of output before operation.

FURTHER ANALYSIS OF THE ETHER CASES

It next seemed wise to analyze the results after ether, according to the time of administration of the drug in its relation to the anesthesia. The cases are grouped into eight classes, as will be noted below, and then the general averages of each group, before and after operation, compared. Throughout this paper it will be observed that the actual difference in percentage of phenolsulphonephthalein excreted before and after operation is not taken as the "percentage of decrease"; to obtain this latter the actual number of points decrease is divided by the output before operation. The results shown by the following figures are striking in that they show a very distinct relation between the time of injection and the output of the drug.

1. Injection just before or during operation:
Number of cases, 16.
Output before operation, 61.7 per cent.
Output after operation, 46.2 per cent.
Difference ..... 15.5 per cent.
Decrease in output, 25.1 per cent.

2. Injection within one hour of end of anesthesia:
Number of cases, 8.
Output before operation, 63.4 per cent.
Output after operation, 47.8 per cent.
Difference ..... 15.6 per cent.
Decrease in output, 24.7 per cent.

3. Injection between one and two hours after anesthesia:
Number of cases, 24.
Output before operation, 65.6 per cent.
Output after operation, 52.0 per cent.
Difference ..... 13.6 per cent.
Decrease in output, 20.9 per cent.

4. Injection between two and four hours after anesthesia:
Number of cases, 125.
Output before operation, 60.2 per cent.
Output after operation, 48.0 per cent.
Difference ..... 12.2 per cent.
Decrease in output, 20.2 per cent.

5. Injection between four and six hours after anesthesia:
Number of cases, 94.
Output before operation, 58.7 per cent.
Output after operation, 47.3 per cent.
Difference ..... 11.4 per cent.
Decrease in output, 19.4 per cent.

6. Injection between six and twelve hours after anesthesia:
Number of cases, 21.
Output before operation, 63.2 per cent.
Output after operation, 52.0 per cent.
Difference ..... 11.2 per cent.
Decrease in output, 17.7 per cent.

7. Injection between twelve and twenty-four hours after anesthesia:
Number of cases, 12.
Output before operation, 55.3 per cent.
Output after operation, 51.4 per cent.
Difference ..... 3.9 per cent.
Decrease in output, 7.0 per cent.

8. Injection between twenty-four and forty-eight hours after anesthesia:
Number of cases, 25.
Output before operation, 59.2 per cent.
Output after operation, 57.0 per cent.
Difference ..... 2.2 per cent.
Decrease in output, 3.7 per cent.

These figures seem to demonstrate that the output of phenolsulphonephthalein is affected most during or soon after operation, and returns to approximately normal in forty-eight hours.

Though the amount of ether used in an operation is by no means always indicative of the state of narcosis of the patient, it was decided to study the cases from this point of view also, though in a good many of the whole series the amount of ether could not be ascertained, and at best it is only approximate. In all of these cases the test was done within six hours of the end of the operation.

1. Amount of ether, 1-4 oz.
   Number of cases, 1.
   Output before operation, 55.5 per cent.
   Output after operation, 50.0 per cent.
   Difference ..... 5.5 per cent.
   Decrease in output, 9.9 per cent.
2. Amount of ether, 4-12 oz.
   Number of cases, 161.
   Output before operation, 59.8 per cent.
   Output after operation, 49.1 per cent.
   Difference ..... 10.7 per cent.
   Decrease in output, 17.8 per cent.
3. Amount of ether, 12-20 oz.
   Number of cases, 73.
   Output before operation, 61.2 per cent.
   Output after operation, 45.8 per cent.
   Difference ..... 15.4 per cent.
   Decrease in output, 25.1 per cent.
4. Amount of ether, 20 oz. or more:
   Number of cases, 17.
   Output before operation, 60.4 per cent.
   Output after operation, 44.5 per cent.
   Difference ..... 15.9 per cent.
   Decrease in output, 26.3 per cent.

These results are interesting and instructive, and if they may be depended on, they show clearly that the effect on the kidney, as shown by phenolsulphonephthalein, is directly dependent on the amount of ether used. These figures are probably the least reliable of any, on account of the difficulty of knowing accurately the amount of ether the patient actually gets.

The ether cases were again interpreted to show the effect of the duration of the operation and the anesthesia. They are divided into three groups, and in every case the second test was done within six hours of the end of the operation.

1. Length of anesthesia, one hour or under:
   Number of cases, 151.
   Output before operation, 61.1 per cent.
   Output after operation, 50.3 per cent.
   Difference ..... 10.8 per cent.
   Decrease in output, 17.6 per cent.

2. Length of anesthesia, from one to two hours:
Number of cases, 95.

| | | |
|---|---|---|
| Output before operation, | 60.8 | per cent. |
| Output after operation, | 46.3 | per cent. |
| Difference ..... | 14.5 | per cent. |

Decrease in output, 23.8 per cent.

3. Length of anesthesia, over two hours:
Number of cases, 14.

| | | |
|---|---|---|
| Output before operation, | 52.2 | per cent. |
| Output after operation, | 38.5 | per cent. |
| Difference ..... | 13.7 | per cent. |

Decrease in output, 26.2 per cent.

SUMMARY

| Duration | No. Cases | Decrease % Phthalein |
|---|---|---|
| 0-1 hr. | 151 | 17.6 |
| 1-2 hrs. | 95 | 23.8 |
| 2 hrs. | 14 | 26.2 |

Here is another fairly striking result — that the length of the operation and anesthesia bears a definite relation to the postoperative excretion of phenolsulphonephthalein.

### CASES SHOWING INCREASED EXCRETION OF PHENOLSULPHONEPHTHALEIN AFTER OPERATION

Of the complete series, eighty-four cases showed a higher output after operation than before. It seems significant that, in spite of the fact, the general trend of results after operation is consistently downward. It appears that most of these cases can be explained in such a way as not to change ones convictions as obtained from the general averages. A more careful study of these eighty-four cases shows that:

1. Forty-nine out of the eighty-four cases showed an increase of less than 10 points. If it be assumed, from figures given above under the heading of normal variations in renal function, that the output may vary from day to day, then it may be said that the majority of these cases which show increased phenolsulphonephthalein output have not been actually stimulated to increased work, but rather have not been affected one way or another by the anesthesia and the operation. If this be agreed for cases showing slight increase, the same would have to be said for all those cases showing a similar amount of decrease.

2. In four of the cases, to wit., (a) subdiaphragmatic abscess, (b) empyema, (c) lung abscess and (d) reduction of a painful fracture, the patient's mental or psychic condition was greatly relieved, as well as his suffering, and this might tend to aid his renal function.

3. In eight of the cases the tests were done over twenty-four hours after the operation.

4. One was a case of gas-oxygen anesthesia.

5. Two were cases of local anesthesia.

6. Five were cases of spinal anesthesia.

7. In ten of the cases the results were such as to suggest strongly a possible error, in that it seemed probable that the bladder was not completely emptied at the time of the first test. This may occasionally have been true of some of the others, but there was nothing about them, save the results, to suggest that such might have been the case.

As examples of these cases of increase which are hard to explain, we cite three:

CASE 1.—Man, aged 50; rodent ulcer of face; No. 164.
Test: May 16; 1 c.c. phthalein; voided in two hours. Urine, 145 c.c.; phthalein, 25.0 per cent.
Operation: May 17, gas-ether, 9 ounces; duration thirty-five minutes. Excision of rodent ulcer.
Test: May 17, three hours after close of operation: 1 c.c. phthalein; voided in two hours. Urine, 325 c.c.; phthalein, 42.7 per cent.

CASE 2.—Man, aged 20; No. 65.
Test: January 7, 1 c.c. phthalein; voided in two hours. Urine, 390 c.c.; phthalein, 41.6 per cent.
Operation: January 9, gas-ether, 13 ounces. Duration, half an hour. Appendectomy.
Test: January 9, 1 c.c. phthalein, three hours and forty minutes after operation. Catheterized in two hours. Urine, 300 c.c.; phthalein, 68.4 per cent.

CASE 3.—Man, aged 34; No. 310.
Test: June 25, 1 c.c. phthalein, voided in two hours. Urine, 190 c.c.; phthalein, 36.2 per cent.
Operation: June 26, gas-ether, 10 ounces. Duration, one hour and fifty minutes. Gastro-enterostomy.
Test: June 26, 1 c.c. phthalein, four hours and thirty-eight minutes after operation; catheterized. Urine, 210 c.c.; phthalein, 67.5 per cent.

On the other hand, in contrast to these one sees a few cases in which, without any evident reason, the phenolsulphonephthalein output after operation goes down practically to zero, and that in cases of strong rugged men who have undergone operations neither long or serious. We quote a few of these as examples:

CASE 1.—Boy, aged 19; general health excellent; No. 416.
Test: November 3, 1 c.c. phthalein, voided in two hours. Urine, 125 c.c.; phthalein, 52.6 per cent.
Operation: November 4, gas-ether, 5 ounces. Duration, thirty minutes. Appendectomy.
Test: November 4, 1 c.c. phthalein, two hours and forty-five minutes after operation; catheterized in two hours. Urine, 85 c.c.; phthalein, 16.6 per cent.

CASE 2.—Young man, aged 18, well developed and nourished.
Test: May 19, 1 c.c. phthalein, voided in two hours. Urine, 130 c.c.; phthalein, 76.9 per cent.
Operation: May 20, gas-ether, 4 ounces. Duration, twenty-five minutes. Incision and drainage submaxillary abscess.
Test: May 20, 1 c.c. phthalein, two hours and twenty minutes after operation. Catheterized in two hours. Urine, 200 c.c.; phthalein, 15 per cent.

CASE 3.—Young man, aged 18, well developed and nourished.

Test: May 12, 1 c.c. phthalein; voided in two hours. Urine, 160 c.c.; phthalein, 76.9 per cent.

Operation: May 13, ether, 6 ounces. Duration, forty-five minutes. Radical cure inguinal hernia.

Test: May 13, 1 c.c. phthalein, forty-five minutes after operation. Catheterized in two hours. Urine, 115 c.c.; phthalein, too low to estimate accurately, about 10 per cent.

CASE 4.—Man, aged 37; well developed and nourished.

Test: April 23, 1 c.c. phthalein; voided in two hours. Urine, 675 c.c.; phthalein, 70.4 per cent.

Operation: April 24, gas-ether, 18 ounces. Duration, one hour and five minutes. Appendectomy.

Test: April 24, 1 c.c. phthalein, two hours and fifty minutes after operation. Catheterized in two hours. Urine, 170 c.c.; phthalein, approximately 7.5 per cent.

No one of these patients had a very long or shocking operation; they were all fairly young and healthy, and there seems no reason to doubt the accuracy of the results. Contrast with these:

CASE 1.—Man, aged 24; well developed and nourished; No. 357.

Test: October 22, 1 c.c. phthalein; voided in two hours. Urine, 250 c.c.; phthalein, 62.5 per cent.

Operation: October 23, gas-ether, 15 ounces. Duration one hour and twenty minutes. Appendectomy; exploration.

Test: October 23; patient unconscious; 1 c.c. phthalein, one hour and ten minutes after end of operation. Catheterized in two hours. Urine, 80 c.c.; phthalein, 62.5 per cent. No change.

CASE 2.—Man, aged 41; well developed and nourished.

Test: July 11, 1 c.c. phthalein, voided in two hours. Urine, 250 c.c.; phthalein, 52.6 per cent.

Operation: July 12, gas-ether, 14 ounces. Duration, 40 minutes.

Test: July 12, 1 c.c. phthalein, two hours and twenty minutes after operation. Voided in two hours. Urine, 120 c.c.; phthalein, 52.6 per cent. No change.

CASE 3.—Man, aged 29; well developed and nourished.

Test: August 1, 1 c.c. phthalein; voided in two hours. Urine, 990 c.c.; phthalein, 58.1 per cent.

Operation: August 2, gas-ether, 6 ounces. Duration, 55 minutes. Clamp and cautery for hemorrhoids.

Test: August 2, patient partly out of ether; 1 c.c. phthalein, one hour and twenty minutes after operation. Catheterized in two hours. Urine, 120 c.c.; phthalein, 57.0 per cent. Decrease, 1.8 per cent.

All the foregoing cases were picked out as examples of three types — those showing marked increase, those showing marked decrease and those showing practically no change at all. And in these particular cases, especially those of the first two groups, there is no adequate explanation of the results — just why there was so much increase or so much decrease is hard to say. They tend to confuse one a good deal, and on studying them, with the general averages above in mind, it seems a fair presumption to explain them on the ground of the personal idiosyncrasy of the patient to ether, just as is seen in the reaction of certain individuals to other drugs.

EFFECT OF SHOCK

Cases of pure shock were much desired for this work, but were very hard to get, as almost every patient was either operated on at once, or else died in a few hours without operation. Only one such case of value came in, and it will be given in detail, as it was very interesting:

CASE 1.—Italian, aged 40, well developed and nourished. September 24, brought to the hospital with fractured femur, fractured ribs, fractured fibula and compound fracture of tibia. Patient in such severe shock that he was left on shock table and later put to bed in ward.

Test: 1 c.c. phthalein; catheterized in two hours. Urine, 215 c.c.; phthalein, approximately 5.0 per cent.

September 25: Fractures reduced under ether.

October 7: Patient in good condition, convalescing.

Test: 1 c.c. phthalein; voided in two hours. Urine, 105 c.c.; phthalein, 58.1 per cent.

This is a striking case, and though the only one of its kind, seems to show that shock *per se* has a great inhibitory action on kidney function (cf. Crile's work on shock). Under this heading the following cases are of interest, though they do not present shock alone.

CASE 2.—Man, aged 56; brain abscess; very sick.

Test: October 8, 1 c.c. phthalein; catheterized in two hours. Urine, 75 c.c.; phthalein, 67.5 per cent.

Operation: October 16, gas-ether, 10 ounces. Duration, one hour and fifteen minutes. Craniotomy for brain abscess.

Test: October 16, patient partly out of ether. In shock and on dangerous list. One c.c. phthalein, 5 hours and 55 minutes after operation. Catheterized in two hours. Urine, 90 c.c.; phthalein, 15.0 per cent.

CASE 3.—Man, aged 35, poorly nourished; osteomyelitis of femur.

Test: October 28, 1 c.c. phthalein; voided in two hours. Urine, 110 c.c.; phthalein, 86.2 per cent.

Operation: October 29, spinal anesthesia; hip-joint amputation.

Test: October 29, patient in poor condition and on dangerous list. One c.c. phthalein, seven hours and ten minutes after operation. Catheterized in two hours. Urine, 75 c.c.; phthalein, 26.0 per cent.

Theoretically, there should not be shock during or after an operation under spinal anesthesia, but the patient's condition was such as to suggest that the effect of the anesthetic had worn off and that at the time of the test he was actually in a state of shock — and that the shock and not the anesthetic was the cause of the great decrease in phthalein output.

CASE 4.—Man, aged 40. Fairly nourished. Cancer of rectum.

Test: February 19, 1 c.c. phthalein; voided in two hours. Urine, 90 c.c.; phthalein, 54.3 per cent.

Operation: February 25, spinal plus ether. Combined abdominal and perineal operation for cancer of rectum.

Test: February 25, patient in very severe shock and unconscious. One c.c. phthalein three hours after operation. Catheterized in two hours. Urine, 65 c.c.; phthalein, 00.0 per cent.

It seems here that the shock and not the anesthetic is responsible.

Test: February 26, day after operation, 1 c.c. phthalein; phthalein excretion, 50.0 per cent.

This demonstrated the rapid return to the normal, or to nearly normal.

The following case is not one of shock, though the patient was on the dangerous list at the time of the second test, and died shortly after.

CASE 5.—Man, aged 47, brain tumor.
Test: May 26, 1 c.c. phthalein, voided in two hours. Urine, 90 c.c.; phthalein, 80.5 per cent.
Operation: May 31, ether 10 ounces. Duration, two hours. Craniotomy for brain tumor.
Test: June 2, two days after operation. Patient semiconscious. One c.c.

The foregoing cases have been cited to illustrate what seems to be a very probable fact, namely, that the general condition of the patients, leaving the anesthetic entirely out of the question, and where there is also no kidney lesion, must have some influence on the amount of phenolsulphonephthalein excreted. In Case 1 above, there was no local kidney injury; the surgical shock alone must account for the low output, and, granted this, it is difficult to differentiate in Cases 2, 3 and 4 between the effect of the anesthetic and the effect of the patient's reduced vitality and low ebb of life — at the time. In Case 5 the test was done so long after the operation that neither the anesthetic nor surgical shock could be considered as of any moment, and here again the factor is the poor general condition and what turned out to be impending death.

### KIDNEY CHANGES AS SHOWN BY URINALYSIS

Of all the cases in this series, 208 had complete urinalysis before and after operation. These examinations were not made from the same specimens which were used for the determination of the phenolsulphonephthalein excretion, as it was deemed best to use these entire, for the sake of greater accuracy. Of these 208 cases, twenty-two, or 10.5 per cent., showed after operation either the presence of albumin when it has been absent before, or else an increase in the amount of preexisting albumin. In this list are not included cases of operation on the genito-urinary tract, in which the sequelae of operation, blood and pus in the urine, would naturally give rise to the presence of albumin. Of the twenty-two cases, seventeen were ones in which albumin appeared when it had been absent before, and five were cases of increase. Compared with the figures given at the beginning of this paper, this percentage of 10.5 is neither high nor low — it falls between the extreme figures of the different authors.

The changes in phenolsulphonephthalein output after operation in these twenty-two cases are briefly as follows:

Two showed no change.
Three showed decreased output of 5 per cent. or under.
Three showed decreased output of 5-10 per cent.
Four showed decreased output of 10-20 per cent.
Five showed decreased output of 20-40 per cent.
Five showed increased output.

In view of these results, it seems that the presence of or absence of albumin after operation, bears no definite relation to the increased or decreased output of phenolsulphonephthalein after operation. Also, there were in these cases, sixty-three which showed a decrease in phenolsulphonephthalein output after operation of over twenty points, in which cases the postoperative urine was normal with no albumin. The albuminuria does not go *pari passu* with the decreased excretion of phenolsulphonephthalein.

### THE IMPROVEMENT OF RENAL FUNCTION AFTER IT HAS ONCE DROPPED

The figures given above to illustrate the variations in phenolsulphonephthalein output according to the time after operation at which the test is done show rather clearly that whatever decrease there is to be will come shortly after the operation, and that the longer one waits, the less likelihood there will be of finding a lowered excretion. In other words, it seems that the function will have returned to normal, or practically normal, within from twenty-four to forty-eight hours. To recapitulate briefly, when the test was done within six hours after the operation, the average decrease varied between 19.4 per cent. and 25.1 per cent. Compare this with those cases in which the tests were performed between twenty-four and forty-eight hours after operation, in which the average decrease is found to be only 2.2 per cent. It is fair to assume that of the cases of this last group numerous ones showed a considerable decrease of function within the first few hours, but that it had returned to normal in the twenty-four or more hours before the test was done.

Following are a few cases illustrating this:

CASE 1.—Man, aged 40 (case quoted above also).

Test: February 19, 1 c.c. phthalein; voided in two hours. Phthalein, 54.3 per cent.

Operation: February 25, spinal anesthesia, plus ether. Carcinoma of rectum.

Test: February 25, patient unconscious and in shock; 1 c.c. phthalein, three hours after operation; catheterized. Phthalein excretion, 00.0 per cent.

Test: February 26—day after operation—patient better; 1 c.c. phthalein; excretion, 50.0 per cent.

This case shows total inhibition of phenolsulphonephthalein excretion up to five hours after operation, but return to nearly normal the next day.

CASE 2.—Man, aged 49; well nourished.
Test: November 2, 1 c.c. phthalein; voided. Excretion, 56.8 per cent.
Operation: November 3, 1 c.c. phthalein, three hours and seven minutes after operation; catheterized. Excretion, 39.3 per cent.
Test: Day after operation, 1 c.c. phthalein, voided. Excretion, 57.0 per cent.

CASE 3.—Man, aged 19. Well nourished. Hernia.
Test: November 7, 1 c.c. phthalein. Excretion, 69.4 per cent.
Operation: November 10, hernia.
Test: 1 c.c. phthalein while patient still unconscious; excretion, 00.0 per cent.
Test: November 11 — day after operation — 1 c.c. phthalein. Catheterized. Excretion, 57.0 per cent.

This is a return to nearly normal in twenty-four hours after a drop to zero.

CASE 4.—Man, aged 37, well nourished. Cholecystitis.
Test: October 8, 1 c.c. phthalein; voided in two hours. Excretion, 66.6 per cent.
Operation: October 9, cholecystectomy.
Test: October 9, 1 c.c. phthalein, six hours and thirty-five minutes after operation. Excretion, 40.3 per cent.
Test: October 10, day after operation, 1 c.c. phthalein; voided. Excretion, 80.5 per cent.

In this case the output has not only returned to normal, but has even increased by 13.9 per cent. on the day following operation.

The following case is interesting, as it was one of spinal anesthesia:

CASE 5.—Man, aged 69; well nourished. Op. litholapaxy.

| | App. Time | One Hour Excretion |
|---|---|---|
| Day before operation..... | 12 minutes | 32.0 per cent. |
| Four hours after operation..... | 16 minutes | 22.7 per cent. |
| Twenty-eight hours after operation | 8 minutes | 44.4 per cent. |

This again shows a rebound to a figure higher than the preoperative one.

### THE EFFECT OF LAPAROTOMIES

The following seventy-five cases are laparotomies which were in the series, not including inguinal and femoral hernias. Ventral hernias in scars and umbilical hernias, however, are included, as they can fairly be called laparotomies. In each case the second test was done within six hours of the end of the operation. This group includes widely differing procedures, from simple exploration or rapid appendectomy to extensive intestinal resections or radical stomach surgery.

Number of cases, 75.
Average output before operation, 59.1 per cent.
Average output after operation, 47.2 per cent.
Difference ..... 11.9 per cent.
Decrease in output, 20.1 per cent.

This is, as might be expected, slightly higher than the general average decrease after ether.

THE EFFECT OF OPERATION ON CANCER CASES

It was thought worth while to analyze this group, which includes all the cases of operation for malignant disease — mostly carcinoma. but with a few cases of sarcoma. The operations were of all sorts.

Number of cases, 24.
Output before operation, 54.1 per cent.
Output after operation, 37.7 per cent.
Difference ..... 16.4 per cent.
Decrease in output, 30.3 per cent.

These figures seem to show two things: first, that in accordance with the patient's more or less reduced condition which results from harboring such a growth, the output before operation is lower than normal; and second, that for the same reason the kidneys show the effect of the ether and operation more than the average.

From our series of cases it would be possible to pick out many isolated ones, each showing some point or other of interest, but that would entail the use of a great deal of space and would not prove anything definitely, so that it does not seem advisable to do so at this time.

## *CONCLUSIONS*

In drawing these conclusions we would state that by the term "renal function" is meant "renal function as shown by the phenolsulphonephthalein test." Also, that it does not seem possible to draw a sharp line between the effect of the anesthetic, and the effect of the operation *per se*, and this must be borne in mind in interpreting the results.

1. The output of phenolsulphonephthalein by the kidneys grows progressively less with advancing years.

2. There is a normal daily variation in phenolsulphonephthalein excretion, influenced by many factors, the normal limits of which cannot be defined.

3. After operations under ether there is a decreased output of phenolsulphonephthalein, this decrease being greater in laparotomies than in the general average, and considerably greater in cancer cases.

4. As a general proposition, the sooner after operation the test is done the greater will be the decrease, and the output will have returned to about normal in from twenty-four to forty-eight hours.

5. The more ether that is used, and the longer the operation, the greater will be the decrease in phenolsulphonephthalein excretion.

6. Certain cases will not show decrease, but either no change or increase. Just which cases do, or will do this, cannot be stated.

7. Certain cases will show great decrease or almost total inhibition of the excretion without discoverable cause. Personal idiosyncrasy may enter into this.

8. Shock will cause great decrease in phenolsulphonephthalein excretion. So also will any condition of much impaired vitality.

9. There is no definite relation between decreased drug excretion and the occurrence of albuminuria after operation.

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# INTESTINAL OBSTRUCTION

## AN EXPERIMENTAL STUDY OF THE CAUSES OF SYMPTOMS AND DEATH *

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This experimental study was undertaken for the purpose of explaining certain apparent contradictions in the deductions drawn from experimental work on the development of symptoms and the cause of death in intestinal obstruction. Although a toxin has not been demonstrated in the blood, most observers believe that symptoms and death are the result of a toxemia. The source of the toxin and its physical and chemical properties have not been agreed on, and the manner by which the toxin is taken into the body has not been made clear.

Without attempting an extensive review of the literature, the views held by certain recent workers will be briefly stated.

Murphy and Vincent[1] (1911) from experiments on cats, believe that symptoms and death in intestinal obstruction were the result of the elaboration and absorption of a toxic substance from the obstructed intestine. The toxic substance was believed to be a non-soluble substance which was destroyed by boiling. They emphasized circulatory disturbance, especially venous obstruction, as a factor in causing acute and early symptoms. The toxicity of the loop content was tested with intestine obstructed for from four to six hours. They further believed that the toxin was the result of bacterial growth.

Hartwell and Hoguet[2] (1912) from experimental study on dogs, concluded that death in intestinal obstruction was due to the absorption of a toxic substance as a result of damage done to the intestinal mucosa. They draw no conclusions as to the source of the toxic sub-

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1. Murphy and Vincent: Boston Med. and Surg. Jour., 1911, clxv, 684.

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stance. It is stated that it may be normally present or may result from stagnation. The severity of the symptoms they believe to be in direct ratio to the extent of the damage to the intestinal mucosa. The loss of body fluids is emphasized as a factor in the causation of death. The toxicity of the content of the obstructed bowel was not studied.

Whipple, Stone and Bernheim,[3] and Davis[4] (1912, 1913, 1914) publish the result of experiments on dogs with intestinal obstruction. They believed that death after intestinal obstruction is due to the absorption of a toxin secreted by the intestinal mucosa, and that this toxic secretion can be derived from the bowel wall without a demonstrable change being present in the intestinal mucosa. The perverted secretion is held to be specific for the duodenum and high jejunum, and the toxin to be a soluble substance which is not destroyed by autolysis or boiling in non-coagulable fluid. The toxic material studied was obtained from simple obstruction of isolated duodenojejunal loops of from thirty-six to seventy-two hours' duration. A characteristic symptomatology and pathologic change following the injection of the toxic intestinal content were described.

To summarize: all these observers believe that death is due to a toxemia. Opinions are in conflict as to the solubility of the toxin and its filterability; also as regards the origin of the toxic substance. In a toxemia, two factors must be present, the production of the toxin and the absorption of the toxin. The severity of the toxemia, that is, the symptoms depend on both factors. In the experimental work referred to, these two factors have not been clearly distinguished. The differences in opinion are not so striking when they are analyzed. For example, the opinion of Hartwell and Hoguet in maintaining that the severity of the symptoms depends on the extent of the damage to the intestinal mucosa is not necessarily opposed to that of Whipple and his co-workers in their statement that the toxin may be developed without great changes in the mucosa, for the reason that damage to the mucosa is essential to the absorption of the toxin as is proven by the fact that the toxin is not absorbed from the normal mucosa. Also the terms "damaged mucosa" and "perverted secretion" may not represent different processes. The chemical and physical characteristics of the toxic material were studied by such different methods that varying statements in regard to these properties are not surprising. Murphy and Vincent worked with the content of a strangulated obstruction of from four to six hours' duration. Whipple, Stone, and

3. Whipple, Stone and Bernheim: Bull. Johns Hopkins Hosp., 1912, xxiii, 159; Jour. Exper. Med., 1913, xvii, 286; Jour. Exper. Med., 1913, xvii, 307; Ann. Surg., 1914, lix, 714.

4. Davis, D. M.: Bull. Johns Hopkins Hosp., 1914, xxv, 33.

Bernheim worked with the content of isolated intestinal loops obstructed for from forty-eight to seventy-two hours without strangulation. That the toxicity and the characteristics of the intestinal content should differ after a longer or shorter period of obstruction is not surprising. Thus, when the problem of intestinal obstruction is reduced to its fundamentals, it seems that there is not such a wide difference between the opinions of the experimental workers, except as to source and the physical and chemical characteristics of the toxic material which is responsible for the symptoms and death, the apparently contradictory statements being more the result of undue emphasis on different factors, and the failure to distinguish clearly between the production and the absorption of the toxin.

In this experimental study we have attempted to determine a proper relationship of the various factors and to reconcile, so far as possible, the differences in opinion, and to determine more definitely the factors which lead to the production of this toxic substance. The experiments were carried out on dogs; the operations were done under complete anesthesia with a careful aseptic technic. All animals which did not die from the toxemia were sacrificed by the administration of chloroform. Only one experiment of each subdivision is described in detail. A sufficient number of observations were made in each instance to satisfy us of the correctness of the conclusion, eighty-eight experiments being performed in this study.

### GROUP I.—CLOSED DUODENAL OR HIGH JEJUNAL LOOPS

To show certain characteristics of the intestinal contents after simple obstruction for from seventy-two to one hundred and two hours, and to define the pathologic changes and symptoms following the injection of this intestinal content into another animal.

EXPERIMENT 35.—Dog. Weight, 8,400 gm. Laparotomy. Heavy tapes tied tightly around duodenum just distal to the pancreatic duct and around the jejunum about 30 or 40 cm. distalward. Gastro-enterostomy just distal to tape around jejunum.

Twenty-four hours: Animal is up. Has not vomited.

Forty-eight hours: Animal lies coiled up in cage. No vomiting.

Seventy-two hours: Animal found dead. Weight after death, 8,000 gm.

*Necropsy.*—The abdominal cavity shows nothing abnormal except the obstructed loop. The remainder of the intestine is not distended. Content of the loop is a thick, paste-like fluid, slightly blood-tinged, and having a foul odor. Microscopic examination of the bowel wall shows a complete loss of mucosa except for a few of the deeper tubules.

EXPERIMENT 48.—(*a*) Dog. Weight, 1,100 gm. Laparotomy. Jejunum sectioned twice at 10 cm. and 50 cm. distal to duodenojejunal junction. The four ends turned in. Intestinal continuity reestablished by lateral anastomosis between distal and proximal stump. Abdomen closed. The operation gave an isolated obstructed loop of jejunum.

Twenty-four hours: Animal is up. Does not look sick.
Forty-eight hours: Animal is drowsy. Lies coiled up in cage.
Seventy-two hours: Dog found dead.

*Necropsy.*—Nothing abnormal found except in abdominal cavity. The peritoneum is injected. There is some free fluid. The entire small intestine is distended, the jejunum more than the ileum. The mucosa of the duodenum and jejunum is swollen and red. The isolated loop is tremendously distended and is discolored. There are several areas which are very thin and one pinpoint perforation through which the loop content is escaping. The contents remaining consist of 75 c.c. of a thick, brown fluid of foul odor.

(*b*) Of the fluid obtained from the loop, 15 c.c. was diluted to 30 c.c. with distilled water, and injected into the peritoneal cavity of a dog weighing 4,400 gm. at 11 a. m.

12 m.: Animal is vomiting.
1 p. m.: Dog looks very sick. Vomiting and fluid stools.
2 p. m.: Animal moribund.
3:30 p. m.: Dead.

*Necropsy.*—The peritoneum is intensely injected. The mucosa of the stomach looks normal. The mucosa of the duodenum beginning sharply about 0.5 cm. from the pylorus is swollen and has a deep red color. The jejunum shows a similar condition. The mucosa of the ileum and large intestine shows a less marked injection. Over the swollen, reddened mucosa of the duodenum and jejunum there is a thick, white exudate which can be lifted off in flakes.

(*c*) Another portion of 15 c.c. of obstructed loop content was diluted to 30 c.c. with distilled water. The container was placed in boiling water for ten minutes, the fluid being actively stirred. There was a heavy granular coagulum formed. The fluid was then boiled vigorously over a free flame for one minute. The supernatant fluid and the coagulum were injected into the peritoneal cavity of a dog weighing 4,700 gm. at 10:30 a. m.

12 m.: Dog is up.
1 p. m.: Animal looks very sick. Has not vomited.
3 p. m.: Animal dead.

*Necropsy.*—On opening the abdomen, there is a small amount of colorless free fluid. Peritoneum slightly injected. The coagulum of injected fluid lies in the omentum. The intestine is not noticeably distended. The mucosa of the stomach is normal. The mucosa of the duodenum and jejunum is swollen and red. The mucosa of the ileum and colon is less injected. Culture made from the abdominal cavity gave no growth.

(*d*) Of the remaining loop content, 40 c.c. was diluted to 80 c.c. with distilled water. Centrifuged at high speed twenty-five minutes. The supernatant fluid was filtered through a coarse clay filter and the filtrate passed through a Berkefeld filter. A culture made from the filtrate showed no growth. Of this sterile filtrate, 30 c.c. (an amount which, taking into account the volume of the residue and the loss from evaporation, represented at least one-half of the material in solution in the diluted intestinal loop content) were given intravenously to a dog weighing 5,300 gm. at 9:45 a. m.

10:30 a. m.: The animal looks sick. Has vomited twice.
4 p. m.: The dog still looks drowsy. No more vomiting.

The following morning, the animal did not eat, but recovered during the day and showed no more symptoms.

(*e*) The residue from the centrifugation of 40 c.c. of the obstructed loop fluid (*d*) was made up to 80 c.c. with distilled water, placed in bottle containing several drops of toluol, and set aside at room temperature for forty days. At this time the fluid had separated into two distinct layers, a bottom layer of an opalescent fluid and a top layer of fat-like substance. The fluid at the bottom was filtered through a clay filter and the filtrate passed through a Berkefeld filter. Culture of the latter filtrate gave no growth. Thirty c.c. of the filtrate was given intravenously to a dog weighing 5,700 gm. at 9 a. m.

11 a. m.: Animal looks sick. Has vomited several times, and has passed two fluid stools.

1 p. m.: Dog looks very sick.

6 p. m.: Animal looks distinctly better.

The following morning the animal was sacrificed. Necropsy showed nothing noticeably abnormal.

EXPERIMENT 36.—(*a*) Dog. Weight, 7,350 gm. Laparotomy. Tape tied around duodenum just below opening of pancreatic duct. Another tape placed around upper jejunum. Gastro-enterostomy just distal to distal tape.

Twenty-four hours: Animal is up. Eats and drinks. Vomited once.

Fifty-four hours: Dog is up. Seems slightly drowsy. Anesthetized and abdomen opened. Peritoneal cavity looks normal. The loop of bowel between the tapes is slightly distended. Peristalsis normal. Abdomen closed.

Seventy-two hours: Dog is up. Eats and drinks.

One hundred and two hours: Dog is up. Responds to attention. Weight, 5,400 gm. Animal anesthetized. Abdomen opened. Peritoneal cavity normal. The obstructed duodenojejunal loop was excised. The duodenal and jejunal stumps turned in. The loop is moderately distended. Peristalsis present. The content of the loop measured 100 c.c. The fluid was a thick, paste-like material of foul odor. In gross the mucosa looked strikingly normal. On microscopic examination the mucosa was edematous, infiltrated with mononuclear and polymorphonuclear leukocytes. The epithelial cells in deeper layers of the mucosa showed very little change; from the tips of the villi they were swollen or completely lost.

(*b*) Of the fluid from the obstructed loop 5 c.c. were injected intravenously into a dog weighing 6,700 gm. at 10 a. m.

10:30 a. m.: Immediately after recovery from ether, the animal began to vomit and pass fluid stools.

12 m.: Animal moribund.

2 p. m.: Dog having convulsions.

2:30 p. m.: Dead.

*Necropsy.*—Immediate. Abdominal cavity contained no free fluid. Intestines seemed full but not distended. The entire small bowel had a bluish color. Peristalsis very marked and the contractions rapid. There were no normally progressing waves. On opening the gastro-intestinal tract, the stomach looked normal. Beginning sharply at the pylorus and extending through the jejunum, ileum, and colon, the mucosa was swollen and of a deep red color. The lumen was filled with blood-tinged mucus. The liver, spleen and kidneys were full of dark blood.

(*c*) The dog from which the obstructed loop was excised made an uneventful recovery and remained quite well for some weeks, when the animal died from perforation of the duodenal stump from a collection of meat bones.

These observations justify the following statements:

1. Dogs with isolated obstructed duodenojejunal loop die without infection of the general peritoneal cavity or any evidence that the cause of death is any other than factors immediately associated with the isolated obstructed loop. The symptoms which follow the operation are fairly constant. For the first twenty-four hours after operation, the condition of the animal is not different from that after any laparotomy. Usually at forty-eight hours, the animal appears drowsy and does not eat and vomits. The animal may rapidly become more apathetic and die in from seventy-two to ninety-six hours.

2. Excessive vomiting or extreme loss in body weight are not necessarily factors in the symptoms as is shown by Experiment 35.

3. That symptoms are not the result of the loss of function of the duodenum and upper jejunum as once held by Matthews, is proved by Experiment 36, in which the symptoms cleared up after total excision of this part of the intestine.

4. The content of the obstructed loop is a toxic fluid which, when introduced in comparatively small doses into the circulation of normal dogs, causes severe vomiting, fluid stools, and death. Associated with the symptoms, there is a characteristic pathologic picture which consists in an acute hemorrhagic enteritis, especially marked in the duodenum and jejunum.

5. The toxicity of the obstructed loop content is not destroyed by heating to 60 C. until sterile, or even by boiling.

6. The toxicity of the fluid is very much decreased by filtration through a Berkefeld filter, so that to produce death a dose of filtrate corresponding to several times the lethal dose of unfiltered fluid is necessary.

7. The amount of filterable toxin is increased by prolonged autolysis.

8. In no case did an animal show symptoms without demonstrable changes in the mucosa.

### GROUP II.—CLOSED LOOPS OF ILEUM

To show that obstructed ileum may cause the same pathologic changes and symptoms as obstructed duodenum or jejunum.

EXPERIMENT 41.—(*a*) Small dog. Laparotomy. Two tapes tied around the ileum at distances of about 10 cm. and 40 cm. from the ileocecal valve. The continuity of the intestinal lumen was reestablished by a lateral anastomosis between segments of the ileum proximal and distal to the closed loop.

Twenty-four hours: Dog is up. Eats sparingly.

Forty-eight hours: Animal is up. Has a diarrhea. Vomited several times during the day.

Seventy-two hours: Found dead.

*Necropsy.*—Abdominal cavity contained a small amount of free fluid. Peritoneal inflammatory signs limited to region of entero-enterostomy. The isolated loop is moderately distended. There is a perforation and the intestinal content is passing into the peritoneal cavity. The stomach is not distended, and the mucosa looks normal. The small bowel is somewhat distended, and the mucosa of the duodenum and jejunum is swollen and red. The lumen of the intestine is filled with a thick, soup-like fluid. Microscopic examination of the isolated loop shows extensive degeneration of the mucosa. Of the content of the loop, 5 c.c. of a thick, paste-like fluid of a foul odor was collected.

(*b*) Of the loop content, 2 c.c. was given intravenously to a dog weighing 4,100 gm. at 11 a. m.

1 p. m.: Animal is lying down. Respiration deep. Looks very sick. Has vomited several times.

2 p. m.: Animal completely prostrate. Has passed several fluid stools containing blood.

3 p. m.: Animal dead.

*Necropsy.*—Abdominal cavity contains no free fluid. Stomach normal. The mucosa of the duodenum and jejunum is swollen and deep red in color. The mucosa of the ileum shows a less marked change. The large bowel contains a large quantity of blood-stained mucus.

This experiment illustrates the fact that obstruction of an isolated loop of ileum may produce symptoms and death in seventy-two hours, and that the content of the obstructed loop is a toxic fluid which, when given intravenously, produces symptoms and pathologic changes identical with those obtained from injection of the contents of obstructed duodenojejunal loops. That isolated obstructed loops of ileum may, under certain conditions, be compatible with life, has been demonstrated by Halsted and will be referred to again in this paper. It is significant in this experiment that examination of the loop wall showed marked nutritional changes.

### GROUP III.—ISOLATED, OBSTRUCTED LOOPS WITH INTERFERENCE TO THE BLOOD-SUPPLY

To show the effect of the circulatory disturbance on the development of symptoms and the formation of the toxin.

EXPERIMENT 80.—Dog. Weight, 6,060 gm. Laparotomy. A tape was tied around the jejunum just below the duodenojejunal junction. Another tape tied 30 cm. distalward. The veins in the mesentery of the segment of bowel between the tapes were ligated. The bowel became immediately swollen and blue. Abdomen closed.

Five hours: Animal vomiting.

Seven hours: Dog looks desperately sick. Vomiting.

Twenty-two hours: Found dead.

*Necropsy.*—Showed the obstructed segment soft, friable. Content of the loop which consisted in a thick, bloody fluid, was emptying into the peritoneal cavity through a perforation. Peritoneum showed very little change.

EXPERIMENT 40.—Small dog. Laparotomy. Thirty cm. of jejunum isolated. Ends turned in. The continuity of the intestinal lumen was reestablished by lateral anastomosis. The arteries in the mesentery of the isolated loop were ligated. The loop was completely wrapped up in omentum.

Seven hours: The animal lies coiled up in cage. Does not respond to attention. No vomiting.

Twenty-one hours: Dead.

*Necropsy.*—The abdominal cavity contains blood-stained fluid. The isolated loop is completely surrounded by omentum. The loop is completely gangrenous. It contains 20 c.c. of thick, blood-stained, paste-like material the usual odor.

EXPERIMENT 84.—Dog. Weight, 4,200 gm. Laparotomy. Four tapes were tied around the upper portion of the jejunum so as completely to occlude three segments of the bowel. The circulation in the proximal segment was not distubed. The veins in the mesentery of the middle segment and the arteries in the mesentery of the distal segment were ligated. The segment with the

venous occlusion immediately became swollen and hemorrhagic. The segment with the arterial ligation became pale. Abdomen closed.

Two hours: Animal is vomiting repeatedly. Looks sick.

Seventeen hours: Dog found dead.

*Necropsy.*—The peritoneal cavity contains approximately 100 c.c. of a thin, blood-stained fluid. No evidence of peritonitis. The segment of bowel without circulatory disturbance appears normal except immediately adjacent to tapes. The lumen contains about 1 c.c. of a mucus-like fluid which is blood-stained in the region of the obstructing tapes. The segment with the venous occlusion is distended, swollen, and dark red in color. The content consists of 40 c.c. of a thick, foul-smelling fluid, the color of a potassium permanganate solution. The segment of bowel with the arterial ligation is distended and yellowish-brown in color. The walls are thin. The contents consist of 15 c.c. of a slightly blood-stained, mucus-like material. With contents of the loops, the following experiments were carried out.

(*a*) Of the contents of the segments with venous obstruction, 10 c.c. were given intravenously to a dog weighing 6,900 gm. at 10:45 a. m.

11:30 a. m.: Vomiting. Looks sick.

3:15 p. m.: Vomiting.

5 p. m.: The animal lies coiled up in cage. Does not respond to attention. Sacrificed. Necropsy showed the peritoneal cavity normal. The lumen of the intestine contains blood-stained mucus. There is a slight injection of the mucosa of the duodenum and jejunum.

(*b*) Of the contents of the segment with venous obstruction, 20 c.c. was diluted to 40 c.c. with distilled water. Centrifuged twenty minutes. The supernatant fluid was filtered through paper and the filtrate again filtered through a Berkefeld filter. Amount of filtrate obtained, 23 c.c. The entire filtrate was given intravenously to a dog weighing 4,850 gm. Following the injection, the animal recovered immediately and did not show any symptoms. Three hours after the injection the animal ate a heavy meal.

(*c*) Of the fluid from the segment with arterial obstruction, 6 c.c. was given intravenously to a dog weighing 5,700 gm. at 11 a. m.

11:30 a. m.: Vomited. Passed stool.

3 p. m.: Vomiting. Numerous fluid stools.

5 p. m.: Dog is moribund. Sacrificed. Necropsy showed the bowel full of blood-stained mucus. The mucosa of the duodenum and jejunum was swollen and dark red in color. The ileum showed less change.

(*d*) In this experiment, the fluid from the peritoneal cavity was not tested, but in other experiments it was found that the fluid accumulating in the peritoneal cavity after strangulation of loops of bowel may be sterile at death, and such fluid may be given intravenously in large doses without producing symptoms.

Three experiments prove the following statements:

1. Animals with obstructed segment of bowel in which there is a disturbance in the blood-supply develop symptoms more rapidly and die in much shorter time than animals with obstructed segments of bowel in which there is no nutritional disturbance.

2. The fluid accumulating in segments of bowel with vascular disturbance, especially arterial obstructions, is very toxic in a period of less than twenty-four hours, at which time an obstructed segment of bowel without vascular disturbance contains practically no content.

3. The Berkefeld filtrate from the contents of an obstructed segment of bowel with venous obstruction of twenty-four hours or less

duration, may be injected intravenously in healthy dogs in comparatively large dose, without causing symptoms.

### GROUP IV.—ISOLATED, DRAINED INTESTINAL LOOPS

To contrast the amount of secretion from the jejunum with that from the ileum, and to show the influence of the difference in the amount of secretion in the production of symptoms.

EXPERIMENT 10.—Dec. 18, 1912: Dog. Laparotomy. The jejunum was sectioned about 10 cm. distal to duodenojejunal junction and again 30 cm. more distalward. The ends of the proximal and distal stumps inverted and the continuity of the intestinal lumen reestablished by lateral entero-enterostomy. The distal end of the isolated segment was inverted and the proximal end pulled out through a stab wound in the flank. Abdomen closed.

Jan. 15, 1913: The animal made a good recovery, and is in good condition. The isolated loop is draining a clear fluid.

Feb. 5, 1913: The animal is in good condition. At 1 p. m. under ether anesthesia the drained end was inverted and the skin closed over.

Feb. 6, 1913: Dog in good condition. Has shown no symptoms.

Feb. 7, 1913: Animal vomited once.

Feb. 8, 1913: Animal is up. Does not eat. Pulse 200. Animal looks sick. At 1 p. m. the incision was opened and the inverted distal end was opened. The isolated loop contained 100 c.c. of a thick, paste-like material having the typical foul odor of the obstructed loops.

March 15, 1913: Following the drainage of the isolated loop, the animal recovered in three or four days to normal condition, and remained in good condition until to-day, when it was noted that the dog was somewhat drowsy and did not eat. The enterostomy has contracted to a small opening 1 mm. in diameter, through which is discharging a small amount of a thick, paste-like materal. Under ether anesthesia, the end of the isolated segment of bowel was pulled out so that the drainage opening was much larger.

May 22, 1913: Following the operation, the animal again became quite normal and healthy. The isolated loop is draining a thin, colorless fluid. To-day the drained end of the isolated loop was again closed by inversion.

May 23, 1913: Animal is up. Does not eat. Vomited once.

May 24, 1913: Dog looks sick. Vomited several times during day.

May 25, 1913: Animal still vomiting.

May 26, 1913: No vomiting. Drowsy. Temperature 102. Pulse 160.

May 27, 1913: Animal in same condition. Laparotomy. Peritoneal cavity normal, except for the isolated segment of bowel which is tremendously distended with a yellowish white fluid. The end of the loop was brought out through a stab wound and left open.

May 28, 1913: Animal found dead. Necropsy showed the isolated loop quite empty. There were areas of hemorrhage and congestion; otherwise abdominal contents normal.

Of the fluid obtained from the loop, a culture showed abundant bacterial growth. Fifteen c.c. of the fluid were given intravenously to a dog weighing 5,200 gm. The animal remained limp and died four hours later. Necropsy showed the abdominal organs tremendously congested. The mucosa of the duodenum and jejunum was swollen and red.

EXPERIMENT 43.—Dec. 5, 1913: Strong dog. Laparotomy. About 20 cm. of the lower end of the ileum was isolated. The intestinal continuity was reestablished by end to end suture. The distal end of the isolated loop was inverted and the proximal end drawn through a stab wound in the flank.

Dec. 23, 1913: Following operation, the animal had a profuse diarrhea which has now ceased. Eats well. Is gaining weight. The enterostomy has contracted until it admits only a small probe.

Dec. 31, 1913: Animal in excellent condition. The enterostomy has closed except for a small pin-point opening through which there is no visible discharge. Pressure on the surrounding tissue, however, caused a small drop of fluid to exude.

Jan. 17, 1914: The dog is in excellent health. No visible drainage from isolated loop. An incision was made by the side of the sinus which seemed to communicate with the lumen of the isolated loop and a tape tied tightly around the bowel. Skin closed over tape.

Jan. 21, 1914: Animal has remained in perfect health. No symptoms. The abdomen was opened and the isolated loop inspected. It has the appearance of quite normal bowel. Peristalsis present. Loop apparently empty. Abdomen closed. Subcutaneous tape which remained tightly tied around the bowel removed.

March 15, 1914: Enterostomy completely closed. Animal healthy.

April 16, 1914: The dog is in perfect health. Very fat. Weight, 16 kg. The enterostomy remains completely closed. Skin over site of drainage perfectly smooth. Laparotomy. The isolated loop was delivered. It was slightly distended, especially at its distal (inverted) end. Peristalsis active. Careful examination showed that the loop was quite isolated and completely closed at both ends. The distal end of the loop was excised and the content removed. The content collected consisted of 35 gm. of a thick, white, paste-like material. The distal end of the loop was inverted. Abdomen closed. A culture from the content of the closed loop gave abundant growth of bacilli and cocci. Microscopic examination showed the material to consist of bacteria, exfoliated epithelial cells, and a granular detritus.

Of the content of the closed loop, 2 gm. were diluted to 10 c.c. with distilled water and injected intravenously into a dog weighing 4,600 gm. at 10 a. m.

10:15 a. m.: Animal prostrate.

11 a. m.: Looks desperately sick. No vomiting. Passed semisolid stool.

12 m.: Animal lies with muscles rigid. Blood-stained mucus passed per rectum.

12:30 p. m.: Dead.

*Necropsy.*—Peritoneum normal. Liver, kidneys, and spleen congested. Entire small bowel has a bluish color. Mucosa of stomach normal. Beginning sharply at the pylorus, the mucosa of the duodenum is swollen and is a dark red color. The mucosa is covered by a layer of mucus. The jejunum shows a similar condition. The injection of the mucosa of the ileum and colon is less marked.

Original animal died third day after turning in loop end.

These experiments illustrate that isolated, drained segments of jejunum differ from isolated, drained segments of ileum in the following manner:

1. Isolated, drained loops of jejunum discharge constantly a relatively large quantity of a thin fluid, while isolated, drained loops of ileum discharge very little.

2. Following the closure of such jejunal loops, the animal develops in the course of from seventy-two to ninety-six hours, symptoms of grave intoxication associated with marked distention of the occluded loop and an accumulation of a toxic content. The enterostomy of an isolated, drained loop of ileum may close and the loop may be completely occluded for long periods of time without signs of intoxication. Such a closure is associated with very little and a gradual distention

of the bowel. The content of such a loop may be very toxic without the animal showing any signs of intoxication. Any operative procedure leading to damage of the mucosa results in symptoms.

GROUP V.—THORACIC DUCT EXPERIMENTS

To prove that the symptoms are due to a toxemia and to illustrate the effect of strangulation or distention of the intestine on the absorption of the toxin.

EXPERIMENT 38.—Large, strong dog. Cannula inserted into the thoracic duct at 10 a. m. Fluid from duct collected in sterile tubes.

11:10 a. m.: Laparotomy. Two tapes tied around the jejunum so as to include about 40 cm. of intestine. All of the veins in the mesentery of the occluded loop ligated. Abdomen closed.

11:20 a. m.: Four minutes after ligation of first vein of mesentery, there was sudden change in the thoracic duct fluid. It became blood-tinged and on microscopic examination, showed numerous red blood-cells.

1:30 p. m.: Animal still alive. Fluid from the thoracic duct is dark-red color. Flowing slowly. Coagulates easily.

3:30 p. m.: Animal died.

*Necropsy.*—Chest normal. Abdomen contains 50 c.c. blood-stained fluid. Culture taken. Fluid saved. The obstructed loop is distended, tense, and filled with a thick, bloody fluid. The remainder of the intestine is contracted. The mucosa of the jejunum and duodenum above the proximal tape is injected and contains a thick, mucus-like fluid which was saved. The mesentery of the obstructed jejunal segment is thickened and hemorrhagic. The lymph-glands at the root of the mesentery are swollen, edematous, and blood-stained.

The fluid from the thoracic duct before ligation of the mesenteric veins, measured 125 c.c.; after obstruction, 150 c.c. Cultures taken from thoracic duct fluid, taken every hour during experiment, showed no growth. Also cultures from the mesenteric lymph glands were negative.

Two medium-sized dogs were not given food for twelve hours, and were used for the following experiments:

(*a*) The fluid from the coagulated thoracic duct before the obstruction of the bowel (kept on ice over night) was pressed out and injected into jugular vein. The animal vomited once five minutes after injection, but showed no more symptoms.

(*b*) The fluid from the thoracic duct after obstruction was gotten in a similar manner and injected into the jugular vein. The animal showed no symptoms.

(*c*) Twenty c.c. of the peritoneal fluid given intravenously. Animal showed no symptoms.

(*d*) Ten c.c. of the content of the segment of bowel above the proximal tape, diluted to 50 c.c. with distilled water, filtered through two layers of gauze. Filtrate (40 c.c.) injected intravenously. During the injection, animal became quite limp. One-half hour later animal had completely recovered and showed no more symptoms.

(*e*) Ten c.c. of the content of the obstructed loop, diluted to 20 c.c. with distilled water, filtered through gauze, injected intravenously. The animal looked sick, and would not eat for twenty-four hours, but did not vomit or pass fluid stools. Complete recovery.

EXPERIMENT 37.—Dog. Weight, 10,800 gm. Laparotomy. Tape tied around duodenum just distal to the opening of pancreatic duct. Another tape tied around jejunum 40 cm. distal to duodenojejunal junction. Gastrojejunostomy distal to distal loop. Abdomen closed. Animal ran the usual course. At the end of ninety-six hours, animal was very toxic. Sacrificed. Occluded loop

was distended and contained 100 c.c. of a thick, paste-like material which was collected and used in the following experiment.

A cannula was inserted in the thoracic duct of a strong dog. The fluid collected in sterile tubes. The abdomen was opened and a segment of jejunum was obstructed between tapes. The content of the ninety-six-hour obstructed loop was injected into the occluded loop, and the veins of the mesentery to the obstructed segment ligated. Following the ligation of the veins, the fluid from the thoracic duct became blood-stained. The fluid was collected for four and one-half hours, at which time the animal died. Amount obtained, 75 c.c.

The fluid was kept on ice over night, the fibrin was separated by squeezing through gauze. The entire filtrate was injected under local anesthesia into the jugular vein of a small dog at 9:30 a. m.

10 a. m.: Animal vomiting.

10:15 a. m.: Passed fluid stool.

12 m.: Animal looks desperately sick.

1 p. m.: Dead.

*Necropsy.*—Chest contains a small amount of free fluid. Lungs are edematous. No evidence of thrombosis or embolism. Peritoneal cavity normal. Intestine markedly contracted in rings. Kidneys, liver and spleen congested. Stomach normal. Mucosa of duodenum and jejunum swollen and red. Mucosa of rectum and colon also injected. Ileum normal.

EXPERIMENT 34.—Dog. Weight, 5,000 gm. Laparotomy. Tapes tied around upper jejunum, from 50 to 60 cm. apart. Gastro-enterostomy distal to distal tape. The animal developed the usual symptoms. Found dead after seventy-two hours. The content of the occluded loop was collected and used in the following experiment:

Medium-sized dog. Cannula inserted in thoracic duct. Laparotomy. Tape tied around jejunum. The bowel was sectioned 40 cm. distal to tape and a large cannula tied in proximal end. Following the section of the bowel, the thoracic duct fluid became bloody, but in a few minutes became colorless again. The content of seventy-two-hour obstructed loop was injected into the isolated segment of jejunum. The cannula in the end of the bowel was then connected with water-bottle elevated four feet above the level of the abdomen. This caused a marked distention of the isolated loop of jejunum and immediately following the distention, the thoracic duct fluid became blood-stained. The loop was kept distended and the fluid collected for six hours, at which time the animal died. The fluid from the thoracic duct became more blood-stained as the experiment progressed. Also five hours after beginning of distention, the content of the distended loop was blood-stained. At death, the distended loop was bluish red in color and quite flabby. Microscopic section showed almost complete destruction of the mucosa.

The fluid which was collected from the thoracic duct, measured 75 c.c. Defibrinated and injected intravenously into dog weighing 3,800 gm. at 9 a. m. Animal recovered from anesthetic immediately.

10:30 a. m.: Passed formed stool.

11 a. m.: Vomited large amount of food material.

12 m.: Animal looks sick, shivering. No more vomiting.

The animal recovered during the afternoon and seemed quite well next morning.

These experiments illustrate the following points:

1. Following occlusion of the veins to a segment of bowel, there is immediately hemorrhage into the lymphatics. There is also very quickly an extensive destruction of the mucosa and hemorrhage into the lumen of the bowel.

2. Similar changes follow rapid distention of the bowel from increased pressure within the lumen.

3. With the introduction of the toxic contents of an obstructed, intestinal loop into the lumen of the bowel and the occlusion of the veins of loop containing the toxic fluid (Experiment 37), the fluid from the thoracic duct contains a toxic substance, which, when given to a normal dog, results in symptoms similar to those following the injection of the toxic loop content. The fact that such symptoms did not follow the injection of the fluid obtained from the thoracic duct after simple obstruction and ligation of mesenteric veins of a segment of normal bowel (Experiment 38), is probably a matter of dosage, for in the strangulated loops of six hours, the concentration of the toxin of the loop content is not great (Experiment 84). Another factor is also to be considered. From the fact that following acute venous stasis there is hemorrhage into lumen of the bowel and into the lymphatics suggests that there is free communication between the bowel lumen and the lymph-vascular system, but that this communication is not as free as it seems is shown by the fact that bacteria do not pass into the circulation as evidenced by contents from the thoracic duct remaining sterile. This suggests that the toxin only passes into the circulation in a soluble form, and the content of a strangulated loop for a period of six hours contains very little toxic substance which will pass through a filter. In the case in which the thoracic duct fluid was collected after strangulation of bowel containing toxic fluid, the content was that obtained after ninety-six hours of simple obstruction, at which time there is more soluble toxic material.

That the symptoms were not so severe following the injection of the thoracic duct fluid in Experiment 34 is also probably explained by dosage, for in this case the circulatory disturbance was not so rapid and the mucous membrane probably remained as a protective filter longer, as evidenced by the fact that the content of the distended loop was not noticeably blood-stained until five hours after the distention.

### GROUP VI.—GALL-BLADDER OBSTRUCTION

To show that this toxin found in the content of obstructed, intestinal loops is not specifically a product of intestinal obstruction, and to show the influence of bacterial action on the production of this toxin in the gall-bladder.

The gall-bladder is a hollow viscus, lined with secreting epithelial cells, and embryologically derived from the primitive intestine. Under normal conditions, it contains no bacteria, and it seemed possible that experimental obstruction of the gall-bladder with and without the presence of bacteria might throw some light on the relation of the bacteria to the production of the toxin in intestinal obstruction.

EXPERIMENT 72.—(*a*) March 14, 1914: Dog. Weight, 10,600 gm. High right rectus incision. Gall-bladder emptied of bile. The cystic duct with cystic vessels ligated. Ten c.c. of blood was aspirated from a small mesenteric vein and injected into the gall-bladder. The gall-bladder was then partially separated from the liver, so as to further interfere with its blood-supply. Abdomen closed.

March 15, 1914: Animal is up. Looks well. No vomiting.

March 16, 1914: Dog eats. Does not seem abnormal. Sacrificed.

*Necropsy.*—Immediate. In the region of the gall-bladder there is a localized collection of blood-stained fluid. The gall-bladder looks gangrenous but has not ruptured. Excised *en bloc*. The contents consist of 6 c.c. of a thick, paste-like fluid. A culture taken gave no growth. Microscopic examination of the gall-bladder showed almost complete necrosis.

(*b*) The entire contents of the gall-bladder immediately after removal were injected intravenously into a dog weighing 10,400 gm. at 10:45 a. m.

11:15 a. m.: The animal is still drunk from ether. No vomiting.

12 m.: Dog seems perfectly normal.

5 p m.: Has shown no symptoms. No vomiting. No fluid stools. The animal was quite well the following day.

EXPERIMENT 71.—(*a*) March 11, 1914: Dog. Weight, 13.5 kg. Laparotomy. Gall-bladder emptied of bile. Cystic duct and vessels ligated. With a syringe, 1 c.c. of duodenal contents aspirated. The material from the duodenum was mixed with 10 c.c. of blood aspirated from a mesenteric vein and injected into the gall-bladder. Abdomen closed.

March 12, 1914: Animal is vomiting. Looks sick.

March 13, 1914: Dog found dead.

*Necropsy.*—No free fluid in the peritoneal cavity. In the region of the gall-bladder there is a small amount of fluid. The gall-bladder is bluish red in color. Very little distended. The content consists of 2 c.c. of a thick, paste-like material. Microscopic examination of the gall-bladder shows almost complete necrosis. The mucosa of the duodenum and jejunum is swollen and red. There is a large quantity of mucus in the intestinal lumen.

(*b*) The entire content of the gall-bladder was given intravenously to a dog weighing 5 kg. at 10 a. m.

10:15 a.m.: Animal is up.

10:30 a.m.: Vomiting. Passing fluid stools.

3:30 p. m.: Animal is having muscle spasm.

5:30 p. m.: Animal is moribund. Sacrificed.

*Necropsy.*—Abdomen contains no free fluid. Spleen, liver and kidneys congested. The mucosa of the duodenum and jejunum is swollen and red. There is a large quantity of frothy blood-stained mucus in the intestinal lumen.

EXPERIMENT 73.—(*a*) March 17, 1914. Dog. Weight, 10 kg. Laparotomy. Gall-bladder emptied of bile. Jejunum aspirated. One drop of jejunal fluid mixed with 10 c.c. of sterile water and injected into gall-bladder. Abdomen closed.

March 18, 1914: Dog has vomited several times.

March 19, 1914: Animal does not eat. Anesthetized with very little ether. Abdominal wound opened. Gall-bladder distended. Dark blue in color. Excised *en bloc*. The content consists of 5 c.c. of a blood-stained fluid having the characteristic odor of obstructed intestinal contents. Abdomen closed. Microscopic examinations of the gall-bladder showed necrosis and infiltration with leukocytes.

(*b*) The entire content of the gall-bladder was made up to 10 c.c. with distilled water. Placed in a thermostat at 60 C. for twenty-four hours. Culture taken gave no growth. The fluid was given intravenously to a dog weighing 6.5 kg. at 10:30 a. m.

11:30 a. m.: Animal is up. Has shown no symptoms.

1 p. m.: Dog is lying down. Looks drowsy. Constantly protruding tongue as if nauseated.

2:30 p. m.: Animal has vomited twice. Looks sick.

3:30 p. m.: Animal still nauseated. Passed one semi-solid stool.

The dog recovered without further symptoms, and seemed quite well the following day. These experiments show:

1. The sterile gall-bladder of the dog can be obstructed and the blood-supply interfered with to the point of complete gangrene without the animal showing noticeable symptoms, and the fluid accumulating in such an obstructed gall-bladder can be given intravenously to healthy dogs without inducing symptoms.

2. Following the obstruction and interference of nutrition of the gall-bladder containing the bacteria of the intestine, the animal shows symptoms identical with those of intestinal obstruction, and the fluid accumulating in the gall-bladder when administered intravenously, even after sterilization of 60 C., produces symptoms and a pathologic picture identical with that following the injection of the content of an obstructed segment of intestine.

GROUP VII.—EXPERIMENTS ON A DOG IN WHICH A DRAINED JEJUNAL LOOP BECAME STERILE

To show the effect of bacteria on the production of the toxin in obstructed intestine and to show that the normal secretions of a draining loop of intestine are not toxic.

Jan. 23, 1913: Strong dog. Laparotomy. Jejunum sectioned near duodenojejunal junction and again 30 cm. below. The continuity of the intestinal lumen reestablished by end to end suture. The distal end of the isolated segment was inverted. The proximal end drawn through a stab wound in the flank and stitched to the peritoneum. Abdomen closed.

Feb. 15, 1913: Animal made an unusually good recovery, and is now healthy. The opening of the isolated segment of jejunum is open and there is a constant discharge of a thin, colorless fluid. A mushroom catheter was introduced into the isolated segment. The loop seemed quite empty. By pushing the catheter in and out, there was a marked increase in the peristalsis and the rate of secretion. The peristalsis waves drew the catheter inward.

Six c.c. of the secretion was collected and injected into the peritoneal cavity of a guinea-pig. The pig showed no symptoms.

Feb. 24, 1913: Animal healthy. Drained loop in same condition. By means of a catheter, 20 c.c. of the loop secretion was collected. Incubated at 37 C. for seventy-two hours. Of the incubated fluid, 5 c.c. injected into peritoneal cavity of a guinea-pig. Guinea-pig dead in twenty-four hours.

The remaining 15 c.c. of fluid heated in thermostat twenty-four hours at 70 C. Incubated twelve hours at 37 C. Heated again twenty-four hours at 65 C. Of this fluid, 5 c.c. were given intraperitoneally to a guinea-pig. Pig dead in twenty-two hours. Culture of fluid injected gave no growth. Culture from peritoneal cavity of pig after death gave a growth.

Oct. 15, 1913: Animal is in excellent health. Enterostomy is open and is constantly draining a thin colorless fluid. Peristalsis active and waves in normal direction. A culture made from some content aspirated from the isolated, drained loop remained sterile.

Oct. 20, 1913: Thirty c.c. of the isolated loop secretion was given intravenously to a small dog. The animal showed no symptoms.

Dec. 4, 1913: Three more cultures made at intervals from the secretion of the isolated jejunal loop have remained sterile. The animal is in excellent condition. Anesthetized. Laparotomy. The isolated loop was delivered and obstructed by a clamp made from two short glass rods covered with rubber tubing. An attempt was made to tie the clamp just tight enough to obstruct the lumen without causing necrosis. Abdomen closed.

Dec. 5, 1913: Animal is up. Eats. Seems quite well.

Dec. 8, 1913: The dog has remained quite well. Has not shown any symptoms. Laparotomy (one hundred hours after obstruction of intestine). On opening the peritoneum, a small amount of a cloudy fluid escaped. Culture from this fluid remained sterile. The isolated loop was delivered. It was found that the obstructing clamp had completely severed the bowel. The ends of the bowel apparently occluded by adherent omentum. The isolated loop of bowel inside of the clamp was not distended. The content consisted of 10 c.c. of a thin, colorless fluid which was carefully collected. Both ends of the severed bowel were inverted. Abdomen closed.

Of the content of the bowel a culture was taken which remained sterile. Five c.c. of the content were given intravenously to a dog weighing 6,250 gm. The injection was done without an anesthetic. The animal showed no symptoms.

Dec. 9, 1913: The dog with the isolated, obstructed loop eats, but looks sick. Has vomited several times.

Dec. 10, 1913: Dog does not eat. Vomiting.

Dec. 11, 1913: Animal looks slightly better. No vomiting. Laparotomy. On opening the laparotomy wound there was an escape of a purulent fluid. Abscess well localized at region of wound. The obstructed jejunal loop is tremendously distended. Bluish red in color. Peritoneal coat friable. Excised *en bloc*. Abdomen closed.

A culture of the content of the loop was made by aspirating through a sterilized area. The content consisted of 100 c.c. of a thick, viscid, blood-stained material with a slightly disagreeable odor.

Of the fluid, 10 c.c. were given intravenously to a dog weighing 4,500 gm. Local anesthesia. At the end of the injection the animal showed a marked respiratory difficulty. This respiratory difficulty increased and the animal died in thirty minutes. Necropsy showed a marked cardiac dilatation. The lungs were almost solid from edema. The bronchi were filled with a frothy material. No evidence of embolism or thrombosis.

To another animal, 5 c.c. was given intravenously. The animal was observed for one and one-half hours, during which time there was a marked respiratory difficulty. No vomiting. No fluid stools. The animal seemed quite well the following day.

The culture from the obstructed loop content gave an abundant growth on agar. The growth was "plated" on agar plates. The only organism which could be grown was a Gram-negative streptococcus. Subsequent intravenous injection in a dog of a broth culture of this organism did not produce symptoms.

The animal from which the obstructed loop was excised made an uninterrupted recovery.

1. The secretion of a well-draining jejunal loop, after the complete healing of the traumatism, incident to a creation by operative procedure, may produce no symptoms when given, injected intraperitoneally in guinea-pigs or intravenously in dogs, in doses larger than the lethal dose of occluded jejunal loop content.

2. After continued drainage a jejunal loop may become sterile, in which case complete occlusion may persist for a period longer than

is consistent with life with similar occlusion of such loops containing bacteria, without the animal showing symptoms and without the accumulation of a toxic content.

3. After infection of the isolated loop, continued obstruction leads to symptoms and the accumulation of a toxic content.

4. It is significant in this case that the toxic content, when given intravenously to a healthy dog, produced symptoms and a pathologic picture different from that seen after intravenous injection of the content of ordinary occluded jejunal loops, and that the bacteria in the infected loop were not those of the normal intestinal tract. The possibility of the toxins produced differing under different conditions will be referred to again in the discussion of the toxic substance obtained from the incubation of excised segments of jejunum in the thermostat.

### GROUP VIII.—TOXINS FROM INCUBATION OF SEGMENTS OF INTESTINE IN THE THERMOSTAT

To show that a toxic substance may be produced by the incubation of an intestinal segment which produces symptoms, death, and the same pathologic changes as are produced by the toxin from an obstructed intestinal loop.

EXPERIMENT 66.—(*a*) From a dog 50 cm. of duodenum and upper jejunum was excised. The lumen of the excised segment was filled with blood and placed in a thermostat at 37 C. and allowed to remain six days. In the autolysis a large amount of gas was formed. The end product was a foul-smelling mass of friable solid tissue, and a reddish-violet liquid. The fluid was separated by filtration through gauze.

(*b*) Of the fluid, 10 c.c. was injected intravenously into a dog weighing 10,000 gm. at 10 a. m.

12 m.: Animal is up. No vomiting. No fluid stools.
12:30 p. m.: Dog is vomiting.
1:30 p.m.: Dead.

*Necropsy.*—Abdomen contains no free fluid. The intestine is in a state of spastic contraction. The stomach contains several round worms. Mucosa normal. The mucosa of the duodenum and jejunum is swollen and has a deep-red color. The mucosa of the ileum is not so markedly injected.

(*c*) Twenty c.c. of the fluid was diluted to 40 c.c. with distilled water. Centrifuged ten minutes. The supernatant fluid was filtered through a coarse clay filter, and then through a Berkefeld filter. Culture of the filtrate remained sterile.

Of the filtrate, 20 c.c. was given intravenously to a dog weighing 7,000 gm. at 10 a. m.

11 a. m.: Animal is up. Has vomited once.
1 p. m.: No more vomiting.
2 p. m.: Animal is vomiting.
3 p. m.: Desperately sick. Incessant vomiting. Numerous fluid stools.
4 p. m.: Dead.

*Necropsy.*—Peritoneal cavity normal. Marked congestion of liver and spleen. The stomach is quite normal. The mucosa of stomach is normal. The mucosa of the duodenum and jejunum is swollen and deep red in color. The ileum and colon show a less marked change. The intestinal lumen contains a large amount of mucus.

(*d*) Ten c.c. of the fluid from the autolyzed loop was diluted to 30 c.c. with distilled water. The mixture was boiled over a free flame ten minutes. A sufficient amount of coagulum formed to preclude the possibility of injecting the material intravenously. The entire material was injected intraperitoneally into a dog weighing 8,200 gm. at 10 a. m.

1 p. m.: Animal is up. No vomiting. No fluid stools.

5 p. m.: Dog has been drowsy during day. No vomiting. No fluid stools.

The following day the animal looked sick. Did not eat. Sacrificed. Necropsy showed the injected coagulum imbedded in the omentum. The peritoneum is injected but shows no evidence of infection. The mucosa of the intestine is quite normal.

EXPERIMENT 67.—(*a*) From a freshly sacrificed dog, 50 cm. of duodenum and jejunum was excised. The excised loop was distended with 50 c.c. of sterile water and placed in a thermostat at 37 C. for seventy-two hours. The autolysis resulted in the production of large quantities of gas, and finally a putrid mass of friable material and a thin, cloudy fluid. The fluid was separated from the larger pieces of solid matter by filtering through gauze.

(*b*) 2.5 c.c. of the fluid was given intravenously to a dog weighing 5 kg. at 10 a. m.

11 a. m.: Dog vomited once immediately after recovering from the anesthetic.

11:30 a. m.: Animal very sick. Marked respiratory difficulty. Died.

*Necropsy.*—Lungs show marked edema. Bronchi full of frothy fluid. No evidence of embolism or thrombosis. Heart dilated, filled with blood. The intestines show exaggerated peristalsis. The duodenal mucosa seems a trifle redder than normal.

To prove that the above death was purely a toxic death, 10 c.c. of the fluid was given to another dog weighing 6.2 kg., intraperitoneally. The animal died in thirty minutes, showing the same symptoms and the same pathologic picture after death.

(*c*) Ten c.c. of the fluid from the autolyzed loop was placed in a thermostat at 60 C. and allowed to remain twenty-four hours. Of this fluid a culture remained sterile. Five c.c. given intravenously to a dog weighing 5,890 gm. at 10 a. m.

12 m.: Animal is up. No vomiting. No fluid stools.

2 p. m.: Dog looks drowsy.

3:30 p. m.: Animal in same condition. No vomiting. No fluid stools. No respiratory difficulty.

The following day the animal had completely recovered.

In Experiment 66, the incubation of a segment of duodenum and jejunum in a thermostat at 37 C. resulted in the production of toxic fluid which, when injected intravenously into a healthy dog, produced symptoms and a pathologic picture identical with that following administration of the contents of an occluded intestinal loop during life. Furthermore, the sterile Berkefeld filtrate acted in a similar manner. The toxic fluid differed from that of occluded duodenojejunal loops in that its toxicity was markedly lowered by boiling.

In Experiment 67, the incubation of a duodenojejunal segment in a similar manner resulted in the formation of a toxic fluid which, when introduced intravenously or intraperitoneally, produced symptoms and a pathologic picture differing from that produced by the administration of the content of an occluded segment of intestine. The toxicity of the

fluid was in this instance markedly changed by heating at 60 C. for twenty-four hours.

These experiments show that the toxic fluid resulting from the autoputrefaction of loops of bowel differs when there is no apparent difference in the condition under which the putrefaction takes place. That the placing of blood in the intestinal loop in Experiment 66, and water in the autolyzing loop in Experiment 67 was not responsible for the difference in the toxic fluid was proven by an additional part of Experiment 67 not described in detail, in which an adjoining segment of jejunum to that excised and distended with water, was excised, filled with blood, and autolyzed the same length of time. The fluid obtained gave the same symptoms and pathologic lesions as that obtained from the loop filled with blood. That the two loops produced a toxic fluid with similar properties suggests that the bacterial flora may be the variable factor.

This characteristic clinical and pathologic picture, seen after the injection of the toxic contents of a loop of obstructed intestine or after the injection of the products of autoputrefaction of an intestinal segment, is identical with that which has been found after the injection of certain ptomain poisons. A preliminary chemical examination, which will be extended, indicates that the toxin may belong to the same group of poisons which was isolated by Faust[5] in his study of the ptomains.

It is also interesting to note that the character of the putrefactive process is different from that seen in the lumen of the bowel after simple occlusion of an isolated loop in the living animal. For example, in the former there is a tremendous amount of gas produced which is never seen in the latter. In case the arteries to an isolated, obstructed loop are tied, gas is formed. We have never seen gas in an occluded jejunal loop in which the veins were tied.

### GROUP IX.—EXPERIMENTS TO TEST THE RELATIVE IMPORTANCE OF THE TWO FACTORS, PRODUCTION OF TOXIN AND ABSORPTION OF TOXIN, IN THE CAUSATION OF SYMPTOMS

EXPERIMENT 85.—Dog. Laparotomy. Tape tied around duodenum below pancreatic duct opening. Another tape tied around jejunum, 20 cm. below duodenojejunal junction. Thirty c.c. of the content of an isolated obstructed loop of jejunum of seventy-two hours' duration injected into the lumen of the occluded segment of bowel. Gastro-enterostomy, distal to distal tape. Abdomen closed.

Twenty-four hours: Dog is up. Does not eat.

Forty-eight hours: Animal dead. Necropsy shows the occluded loop tremendously distended with foul fluid.

5. Faust: Arch. f. exper. Pathol. u. Pharmakol., 1904, li, 248.

EXPERIMENT 86.—Dog. Laparotomy. Two tapes tied tightly around the jejunum so as to include from 30 to 40 cm. of its proximal portion. Veins in the mesentery of obstructed segment ligated. Abdomen closed.

Seven hours: Animal vomiting.

Twenty-two hours: Died.

EXPERIMENT 87.—Dog. Laparotomy. Two tapes tied tightly around high jejunum so as to include between them from 30 to 40 cm. of bowel. Seventy-five c.c. of the toxic content from an obstructed jejunal loop of seventy-two hours' duration injected into the lumen of the bowel between the tapes. The veins in mesentery of the occluded loop ligated. Abdomen closed.

Three hours: Animal vomiting.

Nineteen hours: Animal died.

These experiments show that the introduction of toxic fluid into the lumen of occluded intestinal loops with and without strangulation does not materially decrease the length of life after intestinal obstruction. In Experiment 85, in which several times the lethal dose of toxic fluid for intravenous administration was introduced into the occluded loop, the animal lived forty-eight hours. Animals having the same operation with no toxic fluid usually live forty-eight to seventy-two hours. In Experiment 87, in which a large amount of toxic material was introduced into the lumen of a strangulated loop of bowel, the length of life was only three hours less than in Experiment 86, in which the same operation was done except no toxic material was introduced into the obstructed bowel. Hence in the causation of symptoms, the factors leading to the absorption of the toxin are relatively more important than those of the production of the toxin. This fact has an important bearing on the surgical treatment of intestinal obstruction.

## SUMMARY

Certain of these experiments were repetitions of those of other investigators. They were necessary in order to confirm or refute the previous observations and to form a complete series from which conclusions could be drawn concerning the symptomatology and pathology of intestinal obstruction, as well as the reasons for the conflicting views which exist regarding the origin and nature of the toxin in intestinal obstruction and the way in which it is absorbed.

This study justifies the following conclusions:

1. In intestinal obstruction, the content of the obstructed bowel contains a toxin which, when absorbed in sufficient amounts, produces definite symptoms and pathologic lesions and death.

2. The toxins are the result of bacterial growth. They are not specific for any part of the intestinal tract, and may be formed in the gall-bladder.

3. The chemical and physical characteristics of the toxic substance may vary with the length of time which the obstruction has existed as

well as with the different conditions under which the obstruction occurs.

4. This toxin may enter the circulation by way of the thoracic duct.

5. Death after intestinal obstruction is the result of a toxemia which may be independent of infection of the peritoneal cavity or general circulation.

6. This toxic substance does not pass through a normal mucous membrane.

7. In the production of symptoms the factors which make this absorption possible are more important than the factors which produce the toxin.

8. Interference with the circulation of the obstructed intestine is an essential factor in allowing this abnormal absorption.

9. Simple obstruction of a segment of duodenum or jejunum results in earlier and severer symptoms than similar obstruction of a segment of ileum because the secretion into the lumen of the former leads to rapid distention and circulatory disturbance in the bowel wall.

10. The symptoms and pathologic lesions following the intravenous administration of the contents of a segment of bowel after obstruction are the same as those described resulting from intravenous injection of certain of the ptomain poisons.

11. In the surgical treatment of cases of intestinal obstruction, that part of the intestine with a mucous membrane which has been so damaged as to permit of abnormal absorption should be resected rather than drained.

# A WORKING HYPOTHESIS OF HEMOGLOBIN PIGMENT METABOLISM *

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In the following pages arguments in favor of the view that hemoglobin pigment is produced and decomposed in the liver are presented.

This view was suggested by work which has already been published from this laboratory, and to some extent the evidence in favor of it is drawn from that source. New work has been started on the basis of this hypothesis along several lines, but there are so many questions raised, which circumstances prevent us from attempting to answer, that it seemed better to present what evidence we have at present.

It is claimed that those data which we already possess are harmonized and rendered intelligible by the adoption of this view, and that there is sufficient evidence to justify its acceptance as a working hypothesis.

As it was in the hope that applications to clinical medicine might be found that the subject was approached, special stress has been laid on the possibilities opened up along these lines.

## THE DECOMPOSITION OF HEMOGLOBIN PIGMENT

One of the outstanding features of blood metabolism is its intensity, the rapidity with which it is continually being broken down and rebuilt. We have no means of measuring exactly the rate of hemoglobin decomposition and regeneration, but from the data which we possess it has been calculated that not less than 7 per cent. of the total red blood-cells are daily disintegrated in the body. Each individual red blood-cell, therefore, lives for about fourteen days. What is then its fate?

It would seem that there are special cells distributed throughout the body whose function it is to remove decrepit red blood-cells from the blood-stream. These are endothelial phagocytic cells which may differ considerably in morphology, but are united in possessing certain functional capacities. Thus they take up finely suspended "vital" stains, or colloidal metallic solutions, when these are injected into the blood-stream, and they have some connection with cholesterin metabolism,

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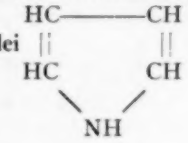
for the doubly refractile droplets characteristic for this substance may be demonstrated within them.

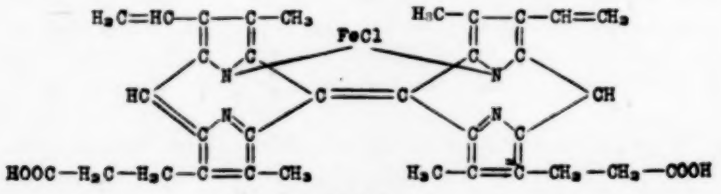
They are situated in the sinuses of the spleen especially, to a less extent within the capillaries of the lymphatic glands and bone-marrow, and stretching across the lumen of the capillaries they constitute in the liver, the Kuppfer or stellate cells.

The cells of this type may be seen to engulf the red blood-cells, and, as the work of Bain[1] and of Paton and Goodall[2] indicates, it is the old and damaged corpuscles which are thus taken up. Within or near the endothelial cell the corpuscle breaks down. There is evidence that in the spleen at least the hemoglobin which is thus set free is excreted into the blood plasma. And the special function of the Kuppfer cells is not only to phagocytose red blood-cells, but also to take up the free hemoglobin which is brought to it in the form of a fine emulsion from the splenic vein. The droplets of hemoglobin have been seen to pass from the Kuppfer cells into the liver cells.

This liberation of hemoglobin from the corpuscles is the first step in the process of hemoglobin pigment decomposition. The next is probably the detachment of the pigment moiety hemochromogen—from the protein globin. Hemochromogen can be readily separated from globin by hydrolysis outside the body, and by the action of hydrochloric acid, a stable substance—hemin—is produced with altering its essential constitution, in which four pyrrol nuclei



are linked to an atom of iron.

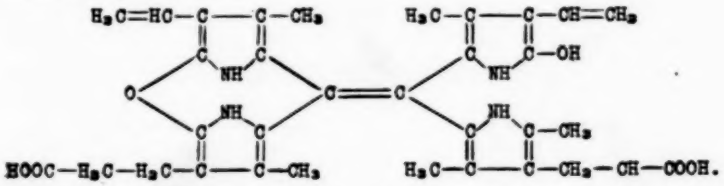



Hemin

The hemoglobin pigment which is thus set free is converted into bilirubin. This is the most important change which it undergoes, but it is the one about which we know least. No one has succeeded in reproducing the necessary reactions outside the body, either in per-

1 Bain: Jour. Physiol., 1903, xxix, 352.
2 Paton and Goodall: Jour. Physiol., 1903, xxix, 411.

fusion, autolytic or digestion experiments, and there is consequently a gap in our knowledge of the chemical processes which are involved. The proof that hemoglobin pigment is the source of the formation of bilirubin rests on experiments which show that when hemoglobin pigment is injected into the blood-stream it produces an almost quantitative increase in the bilirubin excreted in the bile. But although we do not understand the manner in which the conversion is accomplished, we can see the extent and nature of the change by comparing the formulae of hemin and of bilirubin.



Bilirubin

The four pyrrol nuclei are still present, but the iron has gone, and there is more oxygen present.

Where does the change of hemoglobin pigment into bilirubin take place?

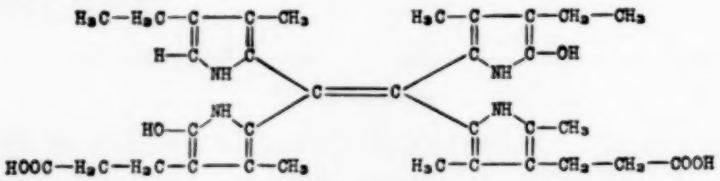
A short time ago there would have been few who would have had any hesitation in maintaining that it was in the liver cell alone that this transformation occurs, although it was admitted that in old blood extravasations a substance indistinguishable from bilirubin was occasionally found.

There is still no doubt that the liver cell under normal conditions is the place where bilirubin is formed. But since the work of Whipple and Hooper[3] and of McNee[4] has appeared, it must be admitted that in all probability, bilirubin can be formed elsewhere, for they have succeeded in producing icterus in the absence of practically the entire liver. This extrahepatic icterus of Whipple and of McNee's experiments finds its clinical counter-part in the cases of hemolytic jaundice in which with no diminution or hindrance to the excretion of bilirubin in the bile, there is, nevertheless, a more or less marked degree of icterus. Widal, some years ago, pointed out that the discarded theory of hematogenous jaundice was the only one which could account for such cases. And, as in Banti's disease, we may reasonably postulate a hyperactivity of the hemolytic function of the endothelial phagocytic cells, so also there is some evidence for the hypothesis that in "hemolytic jaundice" con-

3. Whipple and Hooper: Jour. Exper. Med., 1913, xvii, 593 and 612.
4. McNee: Jour. Path. and Bacteriol., 1914, xviii, 325.

ditions are present which allow this group of cells to assume in part the function of bilirubin formation.

The final stage in the decomposition of hemoglobin is the conversion of bilirubin into urobilinogen. This is an entirely extracellular process which takes place in the intestine. There the bilirubin begins to undergo a change which is the reverse of the process by which it was formed from hemoglobin pigment, in that it is essentially a reduction. Urobilinogen can be made from bilirubin outside the body by the use of strong reducing agents, and its formation in the intestine is no doubt favored by the relative absence of oxidation reactions there, but as yet we know little about the conditions under which it is produced. Hans Fischer and Röse[5] have succeeded recently in isolating it and give the following formula:



**Urobilinogen**

Urobilinogen contains a hydrogen atom attached directly to a carbon atom of one of the pyrrol rings and on account of this grouping, it reacts with paradimethylaminobenzaldehyd to form a red pigment which has a characteristic absorption spectrum.

By making use of this reaction, changes in the amount of urobilinogen in a solution under different conditions can be readily followed. Such studies immediately show that urobilinogen is not an end-product of hemoglobin metabolism. It is an extremely unstable body which readily takes up oxygen, and loses the capacity to form the red pigment with paradimethylaminobenzaldehyd. In its place there appears the pigment urobilin which has certain spectroscopic characteristics which allow of rough estimation.

The studies of Hans Fischer and Meyer-Betz[6] indicate that the formation of urobilin depends on the union of two molecules of urobilinogen under the influence of oxygen, a reaction analagous to the production of indican by the oxidative polymerization of indoxyl sulphates. The reaction may be expressed schematically thus:

Urobilinogen + oxygen + urobilinogen = urobilin.

The reduction of bilirubin to urobilinogen in the intestine is complete. No evidence could be obtained that the conversion stops in man

5. Fischer and Röse: Ztschr. f. physiol. Chem., 1914, lxxxix, 255.
6. Meyer-Betz: Ergebnd. inn. Med. u. Kinderkr., 1913, xii, 733.

at substances intermediary between bilirubin and urobilinogen, although in the dog it would seem probable that this occurs. But the process does not stop with urobilinogen, so that human stools practically always contain a varying amount of urobilin before they are passed. As soon as the stool is exposed to the action of the oxygen of the air, the formation of urobilin goes on more rapidly, especially under the influence of light. This explains the familiar fact that after a stool has stood for some time, it is darker on the surface than in the central unexposed parts. The colorless urobilinogen has been turned into the brown pigment urobilin.

Should we then look on urobilin as the end-product of hemoglobin metabolism? We cannot do so if we define an end-product as a final cleavage product which cannot be made use of or further changed in the body.

Urobilin is not a fixed substance. Its instability has, so far, not allowed of the determination of a constitutional formula, and we do not know with certainty the products of its change. But we do know that the same conditions which favor the conversion of urobilinogen into urobilin also bring about the disappearance of the urobilin itself. I believe that this is a very important fact and that it is the key to the understanding of the whole subject. It will be returned to in considering the formation of hemoglobin.

In summing up our knowledge of the decomposition of hemoglobin pigment we may conclude that, under normal conditions, hemoglobin is liberated from the old red blood-cells by phagocytic endothelial cells; that the hemoglobin thus set free passes into the blood plasma and is taken up by the Kuppfer cells which pass it on to the liver cells. Here the pigment is separated from globin, and after removal of iron and undergoing intramolecular changes which involve the addition of oxygen, is converted into bilirubin. The bilirubin passes into the intestine, and is reduced to urobilinogen, which again in part is polymerized to urobilin.

## THE FORMATION OF HEMOGLOBIN

Is the animal organism capable of forming for itself the keystone in the structure of hemoglobin pigment — the pyrrol nucleus — or is it dependent on the ingestion of preformed pyrrol nuclei in the food?

A new formation of pyrrol within the body has never been demonstrated. That does not mean much, but on the other hand there is nothing inherently improbable in the conception that in the last resort the body has to rely on sources outside of itself for the formation of hemoglobin. We have the analogy of the benzene ring, which can be evolved only by processes peculiar to the vegetable organism, although

it forms an essential part of many of the structures of the animal body (Baumann[7]).

Apart from blood and chlorophyll, the only food constituent which is known to contain the pyrrol nucleus is the amino-acid, tryptophane, a body which is found in considerable amounts in nearly all proteins.

Thanks to the work of Osborne and Mendel,[9] we are in possession of exact and detailed data on the effect of a diet free from blood, chlorophyll and tryptophan on the weight and growth of rats. They show conclusively that under these conditions the body tissues cannot be maintained. There is a progressive loss of weight, which can be at once arrested by the addition of small quantities of tryptophan to the food. They make no mention of anemia, but even if there were no anemia, the possibility of hemoglobin formation by pyrrol synthesis within the body would not be proved, for the total quantity of tryptophan in the body is large, and it is conceivable that with a reduction of the rate of hemoglobin metabolism to a minimum, enough hemoglobin might be formed at the expense of pyrrol liberated from the wasting tissues. This seems to be a rather strained interpretation, however, and it is true that the absence of any note as to pronounced anemia in these observations and in the numerous records of complete starvation experiments, is a point which speaks for the capacity of the body to produce pyrrol for itself, even though it does not by any means prove it. We are planning work in connection with this point, but in the meanwhile it may be left on one side.

So far as the question at issue is concerned, it is enough to know that even if the organism can synthesize pyrrol, this product must of necessity pass through many complicated changes before it is finally welded into the intricate molecule of hemoglobin pigment.

On the other hand, in the urobilinogen in the intestine we have a substance in which the pyrrol nuclei are already linked together in their proper relationships to one another. All that we have learned of the economy of natural processes would tend to make us expect to find that urobilinogen would be used in the synthesis of hemoglobin pigment.

We have proof that a considerable part of the urobilinogen daily formed is not lost to the body by being excreted in the stools, but is absorbed from the intestine in the portal blood-stream. This was proved in Müller's classic experiment where he showed that the urobilinogen which appears in the urine has its origin in the intestine. This has been amply confirmed most recently and most directly by Fischer and Meyer-Betz.[10] With the absence of bilirubin from the

7. Baumann: Ztschr. f. physiol. Chem., 1886, x, 123.
9. Osborne and Mendel: Jour. Biol. Chem., 1914, xvii, 325.
10. Fischer and Meyer-Betz: Ztschr. f. physiol. Chem., 1911, lxxv, 232.

intestine, no urobilinogen is formed, and none can be detected in the urine until bile or urobilinogen are given by mouth. The quantity of urobilinogen and urobilin in the stools is less than the theoretical quantity which should be obtained from the amount of bilirubin entering the intestine. The deficit is absorbed into the blood.

With the recognition of this fact, however, we come to the end of what is generally accepted as established. The suggestion has of course been made that the absorbed urobilinogen is utilized in the formation of hemoglobin, but no hypothesis has been advanced as to the manner of its ultilization or as to its effects on urobilinogen excretion.

Wilbur and Addis[11] not long ago published a paper on "Urobilin" which contained the results of quantitative estimations of urobilinogen and urobilin under physiological, pathological and experimental conditions. Since that paper was written some important work of a purely chemical nature has appeared which throws light on some obscurities and difficulties which were encountered in presenting a theory of urobilinogen excretion.

It is, therefore, now possible to put forward a working hypothesis of hemoglobin metabolism which brings these observations into line, which formulates the relationship between hemoglobin decomposition and formation, and which has a practical application, in that it serves as a theoretical support of a formerly more or less empirical method of recognizing disturbances of liver action.

Urobilinogen is found only in traces in normal urine. But in disease there are two groups of conditions in which large amounts of urobilinogen are excreted in the urine.

In one there is an increased destruction of red blood-cells within the body. Malaria is the typical example. In such cases the proof of an unusual degree of blood destruction is found in the greatly augmented quantity of urobilinogen and urobilin in the stools.

In the other group the amount of urobilinogen and of urobilin in the stools is within the normal limits of variation or is decreased, and there is no evidence of excessive blood destruction. The majority of these cases are outspoken examples of disease of the liver — the initial stages of catarrhal jaundice, the toxemic stage of advanced cirrhosis, suppurative cholangitis, etc. — especially conditions in which the liver as a whole is involved, whereas in such local disease as carcinomatous metastases no very marked urobilinuria may be found. Great increase in urobilinogen excretion is also evident in some cases of acute infections such as lobar pneumonia, where a diffuse liver involvement is probable.

11. Wilbur and Addis: THE ARCHIVES INT. MED., 1914, xiii, 235.

On the whole, the clinical evidence is very strong that the presence of considerable amounts of urobilinogen in the urine is associated with liver disturbance, if marked increase in hemoglobin disintegration can be excluded.

Since the urobilinogen in the urine of these cases of liver disease is known to have its origin in the intestine, the current theory is that the normal liver absorbs the urobilinogen brought to it in the portal vein, whereas the diseased liver allows it to pass into the general circulation from which it is excreted by the kidneys. But in what form does the normal liver absorb urobilinogen? Not as such, for none can be extracted from the liver tissue. Not as urobilin, for this substance also is absent. It does not simply pass through the liver into the bile, for only traces of urobilinogen and urobilin are found there under normal conditions.

It is not even brought to the liver as urobilinogen, for none can be found in the blood, not even when large amounts of urobilinogen are being excreted in the urine.

Obviously, therefore, urobilinogen when it is absorbed is changed into some substance which has not been identified.

It will be remembered that urobilin is formed under the influence of oxygen by the union of two molecules of urobilinogen. But if a solution of urobilin is left exposed to air and light it will steadily decrease in quantity, just as the amount of urobilinogen diminishes when its solutions are left under the same conditions. The parallelism between both phenomena is so marked as strongly to suggest that both are the result of the same process, and that the disappearance of urobilin is simply a continuation of the reaction which is known to underlie the formation of urobilin from urobilinogen, namely, a polymerization. The unidentified substance formed might be called urobilin-complex, and the process might be expressed thus:

Urobilinogen + oxygen + urobilinogen = urobilin.
Urobilin + oxygen + urobilin = urobilin-complex.

The following experiment is an example of many which have been carried out in connection with this point.

An acid alcohol extract of a stool after the addition of zinc acetate in absolute alcohol was found to contain urobilinogen to the dilution value of 17 and urobilin to 20 in each 10 c.c. volume. It was left exposed to the air under the action of diffuse room light. After seven hours samples were removed and separate estimations of urobilinogen and of urobilin were made. The urobilinogen had entirely disappeared, but the urobilin had increased to a value of 40; that is to say, the urobilinogen had been converted into urobilin. But the increase in urobilin would have been still greater if another process had not been

proceeding coincidentally, i. e., the polymerization of the urobilin molecules to form the urobilin-complex. This process we have no means of measuring except by the disappearance of urobilin which is shown in the chart to be progressive, though not regular, since the decrease was more rapid during the day-time under the action of light, than at night.

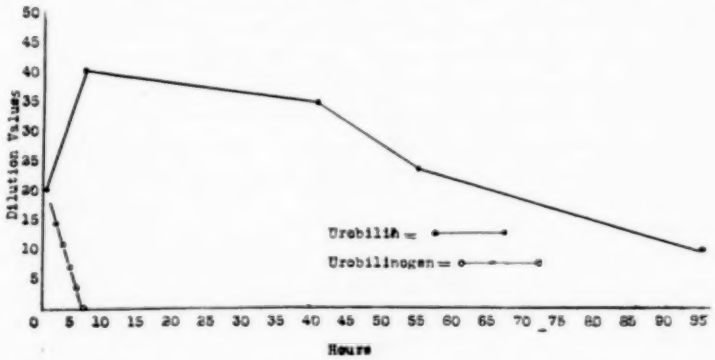



Chart 1.—The dilution values for urobilinogen and for urobilin depend on their respective light-absorbing capacities. Since urobilin has not been isolated its power in this respect cannot be determined with exactitude, and its dilution value must not be assumed to be directly comparable with that of urobilinogen. As a matter of fact, if equal weights of the two substances could be compared, it would be found that the dilution value of urobilin was higher than that of urobilinogen.

In the numerous efforts which have been made to find urobilinogen or urobilin in the blood, nothing has been learned which is in opposition to this hypothesis, and there are some points which indirectly support it.

The blood of patients in whom there is a large excretion of urobilinogen in the urine has never been found to contain any urobilinogen. Urobilin has been found in a few cases. But it is noteworthy that a common factor in such cases is that the patients have been cyanosed. Almost all of them have been cases of lobar pneumonia in the terminal stages with dilated right hearts and marked cyanosis. Now it was shown some years ago by Roth and Herzfeld[12] that when urobilin is added to the blood in vitro it disappears. If it is added to serum alone, however, or even to blood through which carbon dioxid has been passed, it can be recovered. It would seem that both the clinical and the experimental findings may be explained on the assumption that the formation of the urobilin-complex is greatly hastened by oxyhemoglobin, so that it is only when the oxygen of the blood is diminished that urobilin can exist in the blood-stream.

12. Roth and Herzfeld: Deutsch. med. Wchnschr., 1911, xxxvii, 2129.

We may therefore amplify the theory of urobilinogen excretion in the urine of cases of hepatic disease by explaining it as being due to the incapacity of the liver to absorb the urobilin-complex brought to it from the intestine, so that part of this substance escapes past it into the general circulation and reaches the kidneys.

The kidneys excrete only urobilinogen, never urobilin, or urobilin-complex. Probably because of their size, these molecules have to be reduced to urobilinogen before they can pass through the kidney cells. To judge from the prompt appearance of urobilinogen in the urine after injury to the liver, this is a function which the kidneys accomplish without much difficulty, and it would seem reasonable to argue from the fact that only very small traces of urobilinogen are found in the normal urine that the healthy liver is capable of holding almost all the urobilin-complex which is brought to it. The large amounts which may be excreted when the liver is diseased is some indication that the quantity thus absorbed may be not inconsiderable, although it cannot, as we shall see, be taken as a measure of the normal capacity of the liver.

One of the observations of Wilbur and Addis which could not at the time be fully understood was the extraordinary variations in the quantity of urobilinogen in the urine at different periods of the day. Charts 2 and 3 show the quantities of urobilinogen excreted every two hours in a case of bronzed diabetes.

This case was observed over a long period, and at no time was there any relation between the excretion of urobilinogen and of water, urea, chlorids or sugar in the urine. Every night the amount of urobilinogen was greatly decreased as compared with the amount which was passed during the day, but when the patient's habits were reversed and he slept during the day and remained awake and took his meals through the night, the quantitative relationship between the day and night urobilinogen was also reversed. This cannot be satisfactorily explained simply by variations in the absorption of urobilinogen derived from bilirubin, for this is a process which occurs mainly in the large intestine, and there is no reason to believe that there are any sudden changes either in the formation or in the absorption of this urobilinogen.

The study of the urobilinogen and urobilin content of the bile also gave results whose signifience was obscure. In post-mortem bile wide variations were found, from traces too small to estimate to very large quantities, and there was apparently no relation between the amount in the feces and in the bile. In following the daily variations in the urobilinogen and urobilin in the bile discharged from gall-bladder fistulas in patients in whom the gall-bladder had been drained for gall-stones or cholecystitis, it was demonstrated clearly that during the periods

when the patient was feverish and toxemic the quantity of urobilinogen and urobilin was greatly increased, even though little bile was reaching the intestine, and there was consequently little urobilinogen in the stools.

These anomalies in the urobilinogen excretion by way of the urine and bile were not to be explained by any of the current views, and they were simply recorded as facts which any future theory would have to reckon with. The following hypothesis does elucidate not only these facts, but also puts the other data which have been accumulated in their proper place as parts of an ordered sequence of events.

The liver is the central and regulating organ in the metabolism of hemoglobin, not only of its decomposition, but also of its formation. Hemoglobin is converted by the liver into bilirubin. Bilirubin is excreted in the bile into the intestine and is changed to urobilinogen, part of which is excreted in the stools and part absorbed into the portal blood-stream. The urobilinogen in the blood is polymerized to the urobilin-complex. The urobilin-complex is practically all taken up by the liver, but a small amount gets past into the general circulation and is decomposed in the kidneys to urobilinogen and excreted.

But what happens to the greater part of the urobilin-complex which under normal conditions does not get past the liver?

The idea that urobilinogen may be stored in the liver in the polymerized form of urobilin-complex, just as sugar is stored as glycogen, comes at once to the mind as a possibility. Now we know something about the capacity of the liver in storing glycogen, and something also about its power to contain fat, so that it would not be surprising to find that the liver exercised a similar function in connection with hemoglobin metabolism. On the other hand, we also know that the storage capacity of the liver is one of the last of its functions to be disturbed in disease. No matter how disorganized a liver may be, so long as life continues it retains the power to hold glycogen. The relative freedom of the liver from glycogen which is frequently seen post mortem, is the result of diminished intake and increased utilization of sugar, rather than of any inability of the liver to store glycogen, for it is not relatively passive, but active functions which are the first to fail.

But it is preeminently the failure to hold the urobilin-complex which is the first indication of hepatic disturbance. For instance, any toxemic condition, such as that which accompanies an acute tonsillitis, is usually followed by a marked increase in the excretion of urobilinogen in the urine. The liver as a whole is no doubt adversely affected in such conditions, but not as a rule sufficiently to disorganize any of its other functions to an extent which permits us to recognize any abnormality.

Again, it is a well-known fact that if there is need for it the liver can make room for enormous quantities of glycogen, as much as 18 per cent. of the liver weight may be pure glycogen, after large quantities of carbohydrates have been taken.

But even apart from disease of the liver, the limits of its capacity to hold the urobilin-complex are relatively narrow. Since so large a proportion of the total hemoglobin of the body is daily decomposed, it is surprising to find what a relatively small increase in the amount of hemoglobin destroyed is sufficient to lead to the escape of part of the urobilin-complex past the liver.

The conception of a simply passive storage of urobilin-complex in the liver seems to me to be entirely inadequate to account for these facts. It would rather seem that the liver had some active function to perform in connection with the urobilin-complex — a function in which many factors were concerned, which therefore could not be made to work overtime simply by an increase in the urobilin-complex alone, and so intricate that unfavorable circumstances acting on the liver as a whole might readily slow or stop it.

A function which certainly answers this description is the syntheses of hemoglobin-pigment, and the suggestion is put forward that the liver rearranges the atoms in the urobilin-complex to some nearly complete stage in hemoglobin pigment formation in which it is ready for assimulation by the erythroblasts of the bone-marrow.

Such an idea does not find ready acceptance, because one is accustomed to look on the bone-marrow as the site of hemoglobin formation. But so far as I have been able to find the only reason for this belief is the fact that it is possible to trace there all the stages between the early erythroblast, with apparently no hemoglobin, up to the fully formed erythrocyte full of hemoglobin. But that does not prove that the pigment is actually formed within these cells. When hemoglobin is in a coagulated form (Miura[13]) it no longer has its characteristic color or staining reactions, and it may very well be that the erythroblast simply has the capacity to absorb hemoglobin or hemoglobin pigment in some physically altered form, and that the apparent growth of hemoglobin which can be observed within this cell as it develops, is simply a gradual reduction of colorless hemoglobin to the form with which we are familiar.

Looking at the matter simply from the point of view of probability, one would be more ready to believe that such an intricate matter as the synthesis of hemoglobin would be carried out in highly specialized cells such as the liver cells rather than in the undeveloped protoplasm of the

13. Miura: Biochem. Ztschr., 1913, xlix, 137.

early erythroblasts. In this connection it will be remembered that Goldmann's[14] work shows that the early embryo is incapable of producing hemoglobin for itself and has to receive it fully formed from the mother through the placenta, and that in the hen's egg Hugonneng and Morel[15] found a substance which they termed hématovine, which is simply hemoglobin in a slightly modified form. Until the liver is functionating the organism is incapable of synthesizing hemoglobin from pyrrol nuclei.

This conception of the liver as the site of hemoglobin formation is the only one which can account for the unexplained facts which have been mentioned in regard to urobilinogen excretion in the bile and urine.

When this function of the liver is in abeyance the urobilin-complex brought to it cannot be used. It is therefore excreted again into the bile in the form of urobilin and urobilinogen. Thus in cases of gall-bladder fistula in which through bacterial infection the liver was damaged, a large amount of these substances was found in the bile, an amount which bore no relation to the small amounts of urobilinogen found in the stools. On the other hand, when the infection was overcome and the liver recovered, only traces of urobilinogen and urobilin were found, even when enough bilirubin passed into the intestine to lead to the excretion of considerable amounts of urobilinogen and urobilin in the stools. The urobilin and urobilinogen content of the bile depends, therefore, rather on the capacity of the liver to use the urobilin-complex than on the amount which is brought to the liver from the intestine.

There is another point, however, which has to be borne in mind. Urobilinogen is, relatively to urobilin, a readily diffusible substance, and therefore the urobilinogen which is excreted in the bile is in great part rapidly absorbed from the small intestine. In this way the same urobilinogen may continue to circulate between the liver and intestine so long as the liver is incapable of using the urobilin-complex. If in addition urobilinogen derived from the intestinal decomposition of bilirubin is also being absorbed from the large intestine, it can be seen that the quantity of urobilinogen and urobilin in the bile may become very large, as in fact we have found in such cases. Under these circumstances the portal blood-stream will soon become saturated with urobilin-complex and the excretory capacity of the liver be overtaxed, so that part of the urobilin-complex escapes past the liver into the systemic circulation from which it is removed by the kidneys. It is well

14. Goldmann: Aussere und innere Sekretion des gesunden und kranke Organismus., 1912, p. 30.
15. Hugonneng and Morel: Jour. physiol. et de path. gén., 1906, viii, 391.

known that the excretion of bile into the intestine is not constant, but is much augmented at the times when pancreatic secretion occurs. Immediately after the entrance into the duodenum of a bile rich in urobilinogen, there is a rapid absorption of urobilinogen in the form of

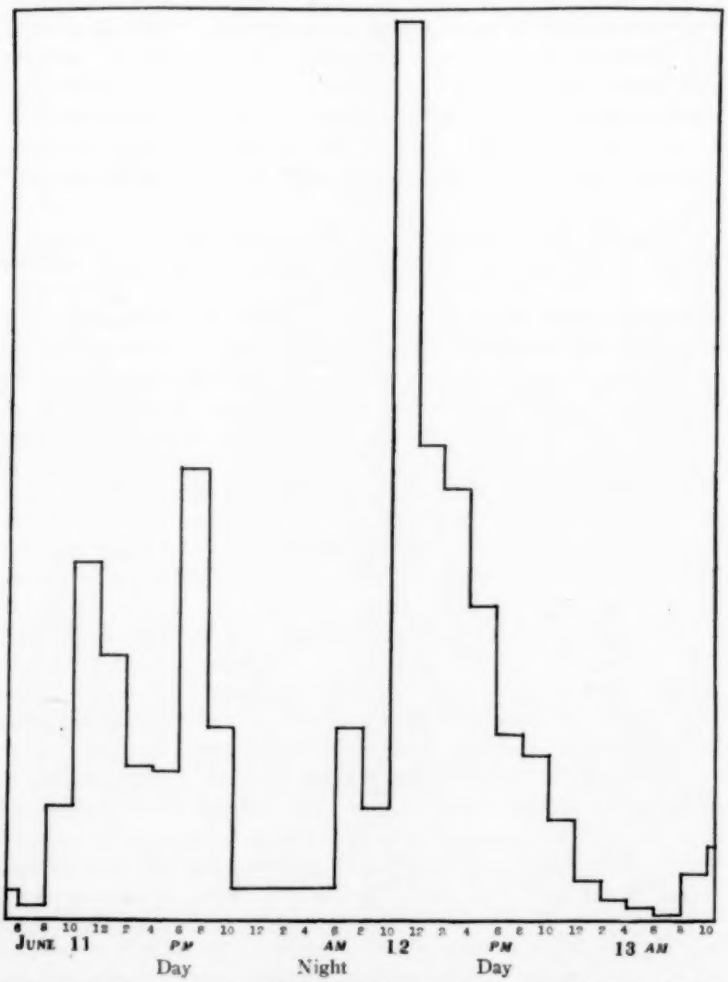



Chart 2.—Excretion of urobilinogen June 11 and 12. Food taken during day time.

urobilin-complex, the liver is overflooded, and as a consequence there is a sharp rise in the urobilinogen excretion in the urine. That the marked excess of urobilinogen excreted during the day is in fact to be

associated with the processes of digestion, is shown by the inversion of this relationship when the food is taken during the night, as is shown in the curves of two-hourly urobilinogen amounts in the case of bronzed diabetes.

The constancy with which any generalized interference with the liver is followed by urobilinogenuria, even though it may be a com-

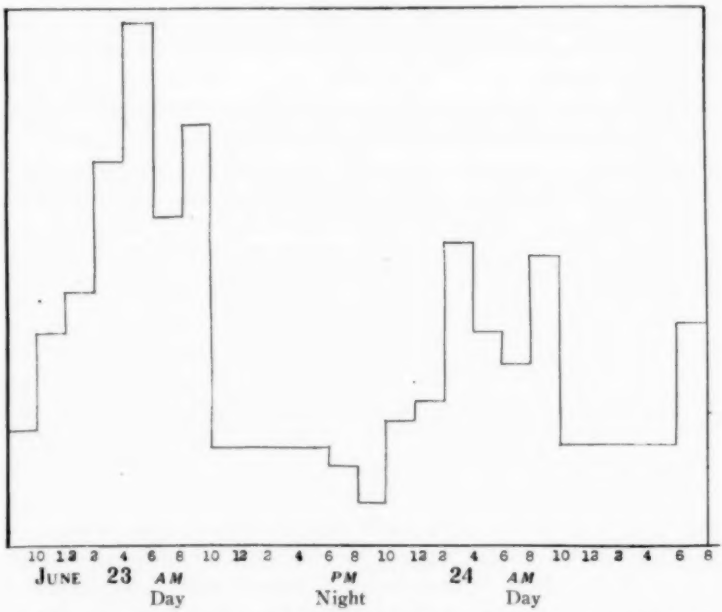



Chart 3.—Excretion of urobilinogen June 23 and 24. Food taken during night. Shows that the largest excretion of urobilinogen occurred during the night instead of during the day, as in the preceding chart.

paratively mild and transient disturbance, and the similar appearance of urobilinogen in excessive amounts in the urine whenever there is even a moderate increase in hemoglobin destruction, is explicable on the theory of a synthesis of hemoglobin from urobilin-complex. It has its analogy in the relative instability of the processes involved in the synthesis of the nitrogen-containing as compared with those concerned with the non-nitrogenous substances in general tissue metabolism. There is a fundamental distinction between protein metabolism and carbohydrate and fat metabolism, which is shown in the fact that when an excessive amount of protein food is taken, the excess is promptly excreted, whereas whatever quantity of carbohydrate or fat is assimilated, is stored in the body. The presence of an excessive amount

of amino-acids does not determine any increase in the reactions involved in the synthesis of new tissue. These reactions require an exact balancing of different constituents, and so any excess of amino-acids above what are required by the tissues is decomposed and excreted as urea in the urine. This parallels the conditions in hemoglobin pigment metabolism, when there has been an excessive formation of bilirubin and as a consequence more of the urobilin-complex is carried to the liver than is necessary for hemoglobin formation. The excess is at once broken down into urobilin and urobilinogen and excreted in the bile.

On the other hand, the flooding of the portal circulation with the urobilin complex, which occurs when the formation of hemoglobin in the liver is in abeyance, has no parallel in the protein metabolism of the tissues. The urobilin and urobilinogen into which the unused urobilin-complex is decomposed as it is excreted through the liver cells, is quickly reabsorbed from the small intestine. With it comes an added increment of urobilinogen from the decomposition of bilirubin. So long, then, as the liver ceases to make use of the urobilin-complex it will heap itself up in the portal circulation, until the blood becomes so saturated with it that the liver can no longer absorb all of it and part escapes into the general circulation to be decomposed in the kidney and excreted as urobilinogen in the urine. In the general protein metabolism there is no such reabsorption of the products of decomposition in order that they may again be used in the building of the tissues, and therefore no accumulation in the blood-stream when anabolism is hindered.

The fact that the hemoglobin-forming function of the liver may show signs of failure, while its other functions, such as, for instance, the capacity to convert sugar into glycogen, are still carried out without difficulty, is explicable when the sensitiveness to any unfavorable change in environment of the comparable synthetic processes concerned in the growth and maintenance of the body tissues, as compared with the relative stability of the general carbohydrate and fat metabolism, is remembered. Bacterial infections, many gastro-intestinal conditions, sometimes even merely psychic disturbances, may lead to a cessation of the maintenance and growth of the protoplasm of the body, even though there are no signs of interference with the power of the tissues to store and utilize carbohydrates and fat. Just as the delicate and intricate reactions on which the building of the nitrogen-containing structures of which the body is composed are the first to be disorganized of all the processes in the general metabolism of the tissues, so in the special metabolism which is carried on in the liver, the rearrangements of the pyrrol nuclei required in the synthesis of

hemoglobin, may be interfered with before the other hepatic functions are involved.

If hemoglobin pigment formation is carried on in the liver and is so easily disturbed, it would follow that any continued interference with this function should be followed by anemia.

It must be borne in mind that here, as in all other important functions of the body, a wide margin of safety may be expected. No doubt the liver is capable, if need be, of producing a much larger amount of hemoglobin than is usually required of it, so that only a fraction of the total liver substance may be sufficient to carry on the work of the whole. In Eck-fistula dogs anemia is not a constant finding, so that it would seem that a relatively small part of the liver may be sufficient if it is functionating normally. It is to be remembered, however, that Enderlen and Magnus-Alsleben[16] have shown recently that the amount of normal liver in an Eck-fistula dog is much greater than is usually assumed, for the hepatic artery rapidly acquires the capacity to give sufficient blood to a great part of the liver. Nevertheless, we know that purely local liver lesions are not necessarily accompanied by urobilinuria and we may assume that so long as a part remains active — how large a part is necessary we do not know — no anemia from decrease in the formation of hemoglobin will result.

The cases in which anemia may be expected are those in which the liver as a whole is involved.

What then are the clinical conditions in which a generalized lesion of the liver exists?

Acute yellow atrophy is not a diffuse lesion. Patches of liver tissue remain and when the patient does not die at once, are sufficiently healthy to grow in size and to divide with great rapidity. Nor is there always any general involvement of the liver cells in conditions grouped under the term cirrhosis, except in the terminal stages.

It is in the cloudy swelling which accompanies severe general infections, such as pneumonia, that one comes nearest to a lesion affecting the liver universally. It is in such cases that one finds large amounts of urobilinogen in the urine, amounts which rise and fall with the course of the disease. It is in such cases, too, that an anemia develops, which becomes more evident the more protracted and severe the infection.

As it is generally understood, the term "anemia" signifies either a decrease in the number of red blood-cells per cubic millimeter of blood, or a fall in hemoglobin percentage, or, as is usual, both combined in varying degree.

16. Enderlen and Magnus-Alsleben: Ztschr. f. d. ges. exper. Med., 1914, iii, 223.

The word, then, covers changes in two substances which under the hypothesis which has been put forward, are genetically distinct, the red blood-cell stroma which is derived from the bone-marrow and its content, the hemoglobin pigment which comes from the liver. Under widely varying physiological and even pathological conditions there is a considerable degree of constancy in any individual in the number of red blood-cells and in the percentage of hemoglobin. Whenever a constant is found we must have a regulating mechanism by which it is maintained. But our methods are as yet too crude to carry us very far in the study of the method of its control. The clinical blood-count and hemoglobin estimation are quite inadequate to give us any idea of the number of red blood-cells and amount of hemoglobin formed and destroyed in a given time, since both are products of two different factors — a balance between production and decomposition. Thus an increase in the number of red blood-cells may mean either that more are being formed or less are being broken up — we cannot tell which. Obviously, we must seek to determine separately the quantity formed and the amount destroyed.

We have no way of directly estimating even approximately the number of red blood-cells or the amounts of hemoglobin which are produced. We can only assume, when young forms of red blood-cells are found, that the bone-marrow is probably active and that the formation of red cells is proceeding at a greater rate than usual.

But we are beginning to get a little insight into one part of the other side of the problem, i. e., the rate of destruction of hemoglobin. We know that the quantity of bilirubin formed is directly dependent on the amount of hemoglobin which is decomposed, and we find that when an excess of bilirubin is excreted a larger amount of urobilin and urobilinogen appears in the stools and vice versa. The proportion of urobilinogen absorbed from the intestine introduces a variable factor, but in spite of this the urobilinogen and urobilin content of the stools gives us a reliable indication of the amount of hemoglobin destruction.

As yet we have not had an opportunity to study more than a few cases of anemia by this method, but the results seem to indicate that there is a reciprocal relation between the destruction and formation of hemoglobin, so that when there is a large amount of urobilinogen and urobilin in the stools, evidences of increased rapidity of production are found, while on the other hand in those cases in which the anemia is due to a deficient formation of hemoglobin, the stool examinations show a decrease below the normal in the urobilinogen and urobilin content, as if the body were endeavoring to bring about a return to the normal by slowing the rate of blood destruction.

This point will be best illustrated by citing three cases in which the hemoglobin percentage was less than normal from three different causes: In Case 1 from increased hemoglobin destruction, in Case 2 from decreased hemoglobin formation because of loss of pyrrol nuclei from the body, and in Case 3 from defective formation because of failure of the hemoglobin-producing function of the liver.

In the following table the normal averages as well as those obtained in these cases are given. The figures for urobilinogen refer to spectroscopic dilution values obtained under the same conditions.

| | Blood | | | Urobilinogen Avg. Daily Excretion | |
|---|---|---|---|---|---|
| | Red Blood Cells | Hemoglobin Per Cent. | Color Index | Stools | Urine |
| Normal ... | 5,000,000 | 100 | 1.00 | 6,475 | 0 |
| Case 1..... | 1,600,000 | 34 | 1.07 | 24,977 | 470 |
| Case 2..... | 2,375,000 | 28 | 0.58 | 2,400 | 0 |
| Case 3..... | 4,100,000 | 65 | 0.79 | 2,095 | 580 |

Patient 1 had pernicious anemia. The figures given above are an average of daily observations for twenty-four days. This time may be subdivided into two periods of fifteen and of nine days, respectively.

| | Blood | | | Urobilinogen Avg. Daily Excretion | |
|---|---|---|---|---|---|
| | Red Blood Cells | Hemoglobin Per Cent. | Color Index | Stools | Urine |
| Period 1... | 1,430,000 | 31-32 | 1.10 | 26,276 | 604 |
| Period 2... | 1,800,000 | 37-38 | 1.04 | 9,546 | 199 |

During the first period the rate of hemoglobin destruction as judged from the excretion of urobilinogen and urobilin in the stools, was more than four times as great as normal. Throughout this period normoblasts and megaloblasts were present in considerable numbers.

During the second period the rate of hemoglobin destruction was only moderately increased. No normoblasts or megaloblasts were found in the smears.

Apparently, therefore, when blood destruction was proceeding rapidly, the rate of blood formation was also increased. That it, however, did not succeed in fully compensating for the increased loss is suggested by the lower hemoglobin percentage and red blood-cell count during this period.

In this case in spite of the marked anemia the corpuscles contained slightly more than the normal quantity of hemoglobin. The reason is not far to seek. There was no failure of hemoglobin-forming power, and the liver was supplied with such an excess of urobilin-complex, that part escaped past it into the general circulation. This, then, is an example of a decrease in hemoglobin percentage due to a preponderance of destruction over formation.

Case 2 was an example of anemia from long continued small losses of blood from internal hemorrhoids. The rate of hemoglobin destruction here was much slower than normal. No evidence of rapid blood regeneration was found. In this case each red blood-cell contained only about half as much hemoglobin as the normal. The production of hemoglobin had lagged behind that of the red blood-cell stromata. Yet there was nothing in the case to support the idea that the hemoglobin-forming power of the liver was failing — no advanced anatomical lesion of the liver, no general infection to cause diffuse liver cell degeneration. That this function was, in fact, intact was shown by the fact that the anemia disappeared after a Whitehead operation had been performed, and the loss of blood had been stopped. Why then, since there was less than the normal amount of blood destruction going on within the body, and since the liver function was potentially, at least, unimpaired, was there any anemia at all? Why did not the body compensate at once for the small losses of blood?

The reason is to be found in the distinction which must be drawn between destruction of hemoglobin within the body, and loss of hemoglobin from the body. In Case 1, although the hemoglobin was destroyed, a considerable part of the urobilinogen into which it was disintegrated was reabsorbed as urobilin-complex and supplied the liver with material for the building of new hemoglobin, whereas when blood is lost from the body as in Case 2 the pyrrol nuclei it contains have gone for good and all. There is no possibility of saving any. If such losses are repeated frequently, even though individually they are small, an impoverishment of the body in pyrrol nuclei may ultimately arise, and even though the liver is fully efficient, hemoglobin formation will fail for lack of building material. Even in those cases in which the loss of blood from the body has not been so great as to lead to a decisive decrease in the pyrrol nuclei available for hemoglobin formation, there may be a relative diminution in the amount of hemoglobin which is produced. For the most powerful stimulus to hemoglobin formation would seem to be the product of hemoglobin disintegration — urobilin-complex. Possibly this substance has some specific excitatory influence on the liver cells apart from its use as raw material in the manufacture of hemoglobin.

Case 3 was in a patient with marked cachexia in whom large, irregular abdominal masses were palpable. An exploratory incision revealed general carcinomatosis.

In this case urobilinogen was found in excess in the urine. There was no increased hemoglobin destruction to account for it as in Case 1; in fact, the rate of hemoglobin disintegration was shown by the stool estimations to be much less than normal. A defect in the capacity to

utilize urobilin-complex because of liver degeneration arising from the poisons which had produced the cachectic state found in the patient, was assumed. As in Case 2, each red blood-cell contained less than its usual quota of hemoglobin. The type of anemia was the same in both cases. They were only to be distinguished so far as the study of the blood and excreta went, by the fact that urobilinogen was present in the urine of the patient in Case 3, and absent in that of the patient in Case 2. But this difference is important, for it points to a difference in causation. The decreased hemoglobin production which was common to both cases was produced in Case 2 by pyrrol starvation, whereas in Case 3 it was derived from an inability on the part of the liver to make use of the pyrrol nuclei with which it was supplied.

The success which has attended the introduction of methods of functional diagnosis in connection with the kidneys, stomach, pancreas, etc., has stimulated many efforts to find a satisfactory test for the liver also.

The functional efficiency of the kidneys can be estimated by the study of the amount and rate of excretion of water, nitrogen, chlorids and phenolsulphonephthalein. By means of the stomach-tube and the Roentgen rays we can arrive at conclusions as to the secretory and motor power of the stomach. Test diets and stool examinations reveal any marked abnormality in the absorptive capacity of the intestine, or in the secretion of the pancreas. But we have no well established and widely used test of liver function. We still have to depend in the diagnosis of most hepatic disorders on anatomical changes which are frequently absent, and on symptoms which are often misleading.

A consideration of the methods of functional diagnosis, which have proved useful in other organs, would indicate that the basis of any hepatic test must be some work performed by the liver alone. Now the liver has many functions, but what are the specific functions peculiar to the liver?

Ten, or even five years ago, the list of such probably exclusive functions was longer and there was less doubt and hesitation about most of them than there is now. Yet it is within these years that great efforts have been made to answer this question. Workers in many different fields have joined in a general attack on the problem from all sides. Clinical pathology has acted as a sort of skirmishing force in the front of this attack, and has often temporarily occupied positions which later have had to be abandoned, as the accumulation of well established knowledge has shown them to be untenable. History, especially recent history, is full of this, and a review of the work of the last decade shows that the advance has been, in the main, a retreat and a retrenchment.

Thus, we used to hold that the synthesis of urea from ammonia, water and carbon dioxid could be carried out only by the liver, but now we know that urea may be produced elsewhere. The more recently advanced test of liver function based on the capacity of the liver to remove ammonia from amino-acids has also been shown, by the fine work of Van Slyke and Meyer[17] and others, to rest on an insecure foundation.

The knowledge that creatin appears in the urine in conditions of disturbed carbohydrate metabolism, and not only in cases in which liver function is particularly disorganized, has thrown doubt on the hypothesis that the transformation of creatin into creatinin is a special function of the liver alone. The work of Fischler[18] with Eck-fistula dogs has shown that Strauss' contention that levulose can be converted into glycogen only by the liver has no foundation in fact, at least as regards warm-blooded animals.

The capacity of the liver to form sugar from certain amino-acids, which may be one of its specific functions, cannot be utilized as a test of liver efficiency except in phloridzinized dogs or the severest forms of diabetes.

The tests founded on the so-called detoxicating functions of the liver have not proved clinically useful and Rothberger and Wintersberg[19] have come to the conclusion that there is no sufficient evidence that the liver has any capacity in this respect which other tissues do not also possess.

In general, it seems that the hopes that have been entertained of finding some distinctive and necessary part played by the liver in the chemical reactions involved in the intermediary protein, fat and carbohydrate metabolism of the tissues, on which to found a functional test, do not at present seem likely to be fulfilled.

The more we learn about the intermediary metabolism of the tissues, the more does it seem that these processes are too generalized to form a basis for testing the function of any one organ.

But there is a group of chemical changes in the body which to a great extent may be regarded as shut off from the general tissue metabolism, namely, the processes concerned with the formation, decomposition, and excretion of the substances which go to form the blood.

It is sometimes said that the blood is a tissue, as much a tissue as muscle or connective tissue. The analogy may be correct in many particulars, but it remains true that the blood is widely separated both

17. Van Slyke and Meyer: Jour. Biol. Chem., 1913-1914, xvi, 213.
18. Fischler: Verhandl. Cong. f. inn. Med., Wiesbaden, 1913.
19. Rothberger and Wintersberg: Arch. Pharmacod., 1905, xv, 339.

in its physical characters and in its functions from any other tissue of the body; and this separation of the blood becomes clearer the more we learn about the chemical changes involved in its metabolism.

Thus the other tissues grow from within themselves by assimilating and incorporating substances abstracted from the blood. But the essential components of the blood are not built within itself, but are given to it ready formed from the other tissues, exactly the reverse process.

The products of the breaking down of the other tissues — urea, uric acid, creatinin, etc. — are removed from them and carried by the blood to the kidneys for excretion, but many, at least, of the special and peculiar end-products of the components of the blood itself are eliminated not through the kidneys, but through the liver.

It is just because of this very separation of the blood metabolism from general tissue metabolism that it is possible to recognize defects in the hemoglobin-forming capacity of the liver. It is this distinction that makes urobilinogen estimations a valuable and practical test of hepatic function. But here the excretion of urobilinogen in the urine alone is not decisive, for a large increase over the usual amount may be determined by augmented hemoglobin destruction quite apart from any liver disturbance. Only when the hemoglobin disintegration has been shown by quantitative estimations of urobilinogen and urobilin in the stools to be within or below the normal limits, does urobilinogenuria acquire any significance as an indication of liver insufficiency.

It may be found that the hypothesis which has been advanced as to the rôle played by the liver in the metabolism of hemoglobin pigment is applicable in some degree to other constituents of the blood. It is significant that the bile acids, taurocholic and glycocholic acid, are most readily accounted for as derivatives of the decomposition of the red blood-cell stromata and that they follow the urobilinogen in its absorption from the intestine and its passage to the liver. The part played by the liver in the formation of fibrinogen has long been known, and it is possible that the serum albumin, serum globulin and the protein constituent of hemoglobin have the same origin.

These are interesting possibilities, but it must be admitted that the data as yet are too meager to allow of any constructive theorizing. It is facts rather that are needed.

It is otherwise with hemoglobin pigment metabolism. Here we have a mass of chemical, experimental and clinical findings which have never been satisfactorily coordinated. In particular, I believe that in urobilinogenuria we have a valuable method of recognizing hepatic disturbances which has been misused and misinterpreted since the time of Jaffé, because the relations between this phenomenon and hemoglobin formation and destruction have never been formulated.

Finally, and this is a point of great importance, if this hypothesis is well founded, we have in urobilinogen and urobilin estimations in the excreta a method which will give us valuable information in the investigation into the cause of various types of anemia.

## SUMMARY

The hemoglobin liberated from outworn red blood-corpuscles is separated into pigment and protein. Within the liver cells the pigment is converted into bilirubin. In the large intestine bilirubin is reduced to urobilinogen.

The change from hemoglobin pigment to urobilinogen is accomplished by intramolecular rearrangements. The four pyrrol nuclei retain their characteristic grouping. Only the side chains are altered.

But in the form of urobilinogen the pigment molecules have acquired two new properties. They have become diffusible, so that they can be absorbed into the portal blood-stream, and they have a tendency to take up oxygen and become linked in pairs, a process of oxidative polymerization which results in the formation of the body known as urobilin.

In the formation of hemoglobin the urobilinogen absorbed from the intestine, since it retains the essential structure of the blood pigment, would form a readily available building material. A difficulty in accepting this as a possibility lies in the fact that urobilinogen has never been found within the body. But here the capacity for polymerization under the influence of oxygen which urobilinogen possesses suggests the reason. The first product of this reaction — urobilin — disappears into some unknown substance, under the same conditions which favor its own formation from urobilinogen. This is assumed to be a continuation of the same process of polymerization, so that a body is formed which consists of an unknown number of urobilinogen molecules linked together. This hypothetical substance has been termed urobilin-complex. It is in this form that urobilinogen exists in the blood and in the tissues.

The kidneys decompose urobilin-complex into urobilinogen. But under normal conditions, although urobilinogen is constantly being absorbed and converted into urobilin-complex in the portal blood-stream, there is practically no urobilinogen in the urine because the urobilin-complex is removed by the liver.

The data we possess on the varying capacity of the liver to absorb and hold urobilin-complex under various experimental and clinical conditions cannot be explained on the assumption that the liver simply stores up the urobilin-complex, but they are in accordance with the conception that the liver has some active work to perform in connection

with this substance. It is maintained that this work consists in the restitution of the original side chains to the pyrrol nuclei of the urobilinogen molecule, so that hemoglobin pigment is formed again from its own decomposition product.

The surest indication of the presence and of the degree of a generalized disturbance of the liver function is based on the determination of the extent of failure of this synthesis of hemoglobin pigment from urobilin-complex.

There is another and more complicated synthesis of hemoglobin pigment from those pyrrol nuclei which are not, as in urobilinogen, prearranged in their proper relationships to one another. For there is a daily loss of pyrrol nuclei from the body in the stools, and the urobilinogen which is absorbed from the intestine cannot replace the total amount of hemoglobin pigment which is constantly being disintegrated. But of this synthesis, of the site, and of the manner of its building, we, as yet, know nothing.

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# A REPORT OF THE BACTERIOLOGICAL EXAMINATION OF ENLARGED LYMPH-NODES REMOVED FROM A PATIENT WITH HODGKIN'S DISEASE*

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The question of the etiological basis of that type of lymph-node hypertrophy referred to as Hodgkin's disease, has for many years been of much interest. There have been two principal ideas in regard to this type of lymph-node hyperplasia; one that it is a neoplasm of the lymphoblastic elements of the lymph-nodes, the other that it is an inflammatory reaction accompanying or following an infection with some micro-organism which attacks especially the lymph-nodes.

The advocates of each of these theories have referred to the histological findings in the lymph-nodes as evidence in support of one or the other of the two theories referred to above. Among others, Gibbons,[1] has expressed the opinion that the enlarged nodes belong in the group of malignant neoplasms. Opposed to this view are McCallum, Longcope, Reed and others who have claimed that the histological findings in the lymph-nodes are those associated with a chronic inflammatory irritant of some kind. Professor Adami in his recent text-book expresses the belief that the lymph-node lesions in Hodgkin's disease are not those of a true neoplasm, but are of a chronic inflammatory nature.

There have appeared in the comparatively recent literature reports that deal with the demonstration of micro-organisms in the nodes themselves. Fränkel and Mutch have reported the results of their study of the enlarged lymph-nodes from a number of cases of "Hodgkin's disease." These authors were able to demonstrate in twelve out of thirteen cases the presence of micro-organisms in the excised lymph-nodes. These organisms were demonstrated in the material remaining after treating the tissues with antiformin. From the morphological and staining characters of the organisms found they were of the opinion that they were either a type of tubercle bacillus or that they belong to a type closely related to the tubercle bacillus.

The infectious basis of Hodgkin's disease has not been limited to the microscopical demonstration of organisms in the lymph-nodes. Negre and Meerimet report the isolation of an organism from the

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* From the Pathological Laboratory of the Montreal General Hospital.
1. Gibbons: Am. Jour. Med. Sc., 1906, cxxxii, 692.

enlarged lymph-nodes removed from two patients with Hodgkin's disease. These organisms were recovered by the use of special medium. Their article gives a description of the organism isolated. The most recent work dealing with the etiology of this disease has been done by Bunting and Yates[2] and reported recently. In their first paper they report the results obtained from the bacteriological study of a series of enlarged lymph-nodes removed from patients whose clinical picture corresponded to that found in Hodgkin's disease, and whose lymph-nodes themselves showed the histological picture found in this disease. The general summary of the work on which this paper is based is as follows: From three out of five cases of Hodkin's disease, in which a bacteriological examination had been made, a pure culture of a diphtheroid, pleomorphic organism has been isolated in pure culture. In the remaining two cases this organism was recognized, but not recovered in pure culture. The morphology, cultural and staining reactions of the organism recovered from one patient is briefly as follows:

> They are facultative anaerobes, Gram-positive, not acid fast, and vary greatly in their morphology depending on a number of factors, e. g., their age, the medium employed and the fluid content of the medium. Pleomorphism is a pronounced feature, coccoid forms, long forms, beaded forms, club-shaped involution forms, are present. Branching forms are present especially on the egg medium. On certain media polar staining forms are present. Plate cultures show rounded, glistening, gray colonies which at the end of twenty-four hours gradually become opaque white. There is a central dark spot in the colony and a fine stippling of the growth.

Since the publication of the above article these same investigators have reported[3] the results thus far obtained by inoculating animals with the diphtheroid organism they have isolated.

They have been able by repeated inoculations not only to infect monkeys (*Macacus rhesus*), but these animals have shown a blood-picture typical of that of early Hodgkin's disease and later have developed chronic lymphadenitis that corresponds grossly and microscopically with Hodgkin's disease in man, of the same duration. Not only have they succeeded in producing these gross and histological changes in the monkey, but they have recovered from the enlarged lymph-nodes a diphtheroid organism similar in every way to the organisms injected. In one of their articles they say:

> Thus, since our experiments demonstrate that the diphtheria organism is pathogenic for monkeys, that it produces a progressive enlargement of the lymph-nodes, with lesions similar to those of Hodgkin's disease in man, and further that the blood changes in the monkey are similar to those in man, we feel fully assured of the etiologic relationship of the diphtheroid organism (*Bacterium hodgkini*) to Hodgkin's disease.

2. Bunting and Yates: Culture Results in Hodgkin's Disease, THE ARCHIVES INT. MED., 1913, xii, 236.

3. Bunting and Yates: Jour. Am. Med. Assn., 1913, lxi, 1803.

We have recently had the opportunity of making a bacteriological study of the enlarged lymph-nodes from a patient with Hodgkin's disease. This opportunity was made possible through the kindness of Dr. J. M. Elder, on whose service the patient was admitted to the Montreal General Hospital. The clinical history is briefly as follows:

*History.*—The patient is a white woman, aged 21. The present illness began two years ago, in 1912, with swelling in the right side of the neck. The swelling increased slowly and did not produce discomfort. In 1913, one year after the first symptoms were noted, a swelling appeared in the left side of the neck. About this time she had a normal pregnancy, after which she became gradually weaker, feverish and easily tired. On exertion she had dyspnea and palpitation of the heart. No pain was present. Patient thinks she lost about fifteen pounds in weight.

Patient has been married five years and has had two children (both died in infancy). She has always been well previous to present illness. There is no family history of tuberculosis.

*Examination of Glandular System.*—At the base of the neck there are two large masses on either side of the middle line. These masses are about 7 cm. in diameter. The skin is freely movable over them. The masses themselves are movable on the underlying tissues and on palpation are found to be made up of discrete individual masses varying in size from 1.5 to 4 cm. in diameter. The smaller growths have a distinctly elastic feel. There is one slightly enlarged lymph-node in the right axilla; otherwise the axillae are free. Below the right clavicle the superficial veins of the thorax are dilated and tortuous. This finding was interpreted as evidence of pressure on the mediastinal vessels. A roentgenogram shows a definite shadow in the mediastinum which corresponds to an area of dulness noted on percussion.

The edge of the spleen is just palpable. On percussion splenic dulness is 16 by 9 cm.

Blood examination shows the following:

| | |
|---|---|
| White cells | 14,200 |
| Red blood-cells | 2,440,000 |
| Hemoglobin | 60 per cent. |

Differential blood-count is as follows:

| | |
|---|---|
| Polymorphonuclear leukocytes | 50 per cent. |
| Small mononuclears | 31 per cent. |
| Large mononuclears | 9 per cent. |
| Eosinophils | 9 per cent. |
| Mast cells | 1 per cent. |
| Blood-pressure | Systolic, 100; diastolic, 80 |

On January 26, 1914, Dr. Elder removed, under local anesthesia, three enlarged lymph-nodes from the left side of the neck. This operation was done with the greatest of aseptic precautions. The three nodes were at once taken to the laboratory for examination. Under careful aseptic precautions a portion of each node was taken for bacteriological investigation, and at the same time sections of the nodes were put into fixing solutions for histological study.

*Pathological Report.*—M. G. H., S-14-58. The specimen consists of three discrete, encapsulated lymph-nodes. They measure 2 cm., 25 cm. and 4 cm. in diameter, respectively. The nodes are loosely united by delicate fibrous tissue. Each node is enclosed in its own capsule and is only loosely united with those about it. The tissues immediately surrounding the lymph-nodes are edematous and contain numerous small blood-vessels. The nodes are only a little firmer than normal. On section they are of a uniform pale, grayish-white color, with a slight yellowish cast. The incised surfaces show a semitranslucent appearance. The following microscopical description is based on Zenker and formalin fixed tissues, sections from which have been stained with eosin and methylene blue, hematoxylin and eosin, Mallory's phosphotungstic acid, hematoxylin, and connective-tissue stain. Specimens from each node have been examined for bacteria both by means of direct smears made from the nodes and by means of staining sections for bacteria. Histologically the sections of the lymph-nodes show briefly the following: The pericapsular tissues are edematous and show a slight cellular infiltration. The infiltrating cells are for the most part lymphocytes. The capsules of the nodes are slightly thickened and infiltrated with lymphocytes. The lymph-node parenchyma has largely lost its normal histological characters. There are no germinal centers. There is a moderate, though not extensive, increase in connective tissue. This connective tissue increase is more marked in certain places. The most striking cellular change is the increase in endothelial leukocytes and the presence of large multinucleated cells. These multinucleated cells occur irregularly distributed throughout the section. There are but few eosinophils found. No necrotic areas or areas of acute inflammatory change are found.

In the direct smears from the nodes there are a few Gram-positive bacilli with morphology similar to the organisms recovered by means of culture. The organisms are few in number. Many fields are examined before one is found. None of them are intracellular. In sections from the nodes stained by Gram-Weigert's method and a modified Gram-Weigert's there are only a very few bacilli demonstrated. These organisms have always appeared as single organisms, they lie free in the tissue spaces and the cellular characters of the tissue immediately surrounding them is in no way different to that elsewhere in the nodes. A careful search for organisms within the cells, especially within the multinucleated cells, has been made. In no instances have we been able to find intracellular organisms.

The bacteriological procedure consisted in excising small bits of each of the lymph-nodes. Some of these were thoroughly macerated in warm sterile saline solution. Other pieces of tissue were inoculated directly in various mediums, the tissue first being thoroughly rubbed over the surface of the medium. The finely macerated tissue was inoculated in large and small amounts on various mediums. Both aerobic and anaerobic cultures were made and incubated at 37.5 C. The inoculated media were examined from time to time. At the end of eighteen hours one tube of human blood agar showed a growth of Gram-positive cocci in groups. None of the other mediums showed positive results until ten days after their inoculation, when a stained smear from one of the hydrocele agar tubes showed Gram-positive organisms. The organisms were mostly of the coccoid and cocco-bacillary type, though a few bacilli were present. Within the next two weeks, twenty-one days after their original inoculation, a number of the tubes showed organisms on stained smears. The presence of colonies on the original tubes could be definitely made out only on certain mediums. Growth was first recognized in the gross by a finely stippled-like appearance to the surface of the medium. The colonies were largest on the hydrocele agar. On this medium they developed near the water of condensation as small, almost transparent colonies. The surface of Dorset egg medium showed a fine stippled appearance. There slowly developed a grayish precipitate in bouillon, and in the water of condensation of the solid medium, especially where the water of condensation was abundant. In two of the tubes of hydrocele

agar, where large amounts of hydrocele had been added, a grayish cloudy growth developed at the bottom of the tube and gradually spread outward between the agar and the inner surface of the culture tube. In the original tubes of solid meduim, growth was most abundant in those which contained large amounts of fluid. The presence of the pleomorphism of the organisms found in our original culture led us to the strong suspicion, at one time, that we were dealing with at least two different organisms. But by transplanting all colonies that showed few bacilli and many coccoid and coccobacillary forms on proper mediums, especially Dorset egg medium, bacillary forms predominated especially in the younger cultures.

A series of transplants has been made on various mediums and the organisms grown through several generations. The morphological characters of these organisms have varied greatly on different mediums, and under varying growth conditions. Cultures of the same age on various mediums show certain characters common to them all and certain characters that vary in the different mediums. They are all Gram-positive and non-motile and none of them is acid fast. The resistance to acid decolorization has been studied by using Gabbet's method and the Ziehl-Neelson method of staining for tubercle bacilli. In using the Ziehl-Neelson method varying dilutions of acids have been employed as decolorizing agents, and the results obtained compared with a young culture of tubercle bacilli (human type), *Bacillus typhosus* and *Staphylococcus pyogenes aureus*. These organisms were smeared on the same slide, and, in so far as possible, smears were made of uniform thickness. In the staining process each slide has been treated similarly. The following results have been obtained: Dilutions of nitric acid for decolorizing were employed from 30 per cent. strength down to 0.5 per cent. The organisms under consideration, *Bacillus typhosus* and *Staphylococcus aureus* were completely decolorized in all of the dilutions used, while the tubercle bacillus remained acid fast throughout.

The morphological characters of the organism in Dorset egg medium is as follows: Most of the organisms are of a definite bacillary type. Many of them show sharp, deeply staining granules within them. These granules may be either unipolar, bipolar, or bipolar with two or more deeply staining areas in the bodies of the organisms between the two poles. The form of the bacillus is not constant. Most of them are comparatively long and narrow and show varying degrees of curving. There are a few branching forms with or without polar bodies in them. As many as four branches have been found in one organism.

A series of roll cultures were made using agar with various percentages of fluid medium, as hydrocele, defibrinated blood, etc.

A series of these tubes was inoculated under both anaerobic and aerobic conditions. In all the roll cultures growth developed, but the most luxuriant growth took place in the anaerobic cultures, especially in those containing proportionately large amounts of fluid. In both the anaerobic and aerobic cultures colonies did not appear at all or only sparsely and slowly within the first 1 to 1.5 cm. of the upper surface of the medium. At this point there developed a narrow band of closely packed colonies. This band varied in width: in some of the tubes it reached 3 cm. in others it was only a few millimeters. Below this point the colonies were not nearly as numerous though they were present throughout the remainder of the mediums. There slowly developed a heavy grayish diffuse growth between the outer surface of the medium and the inner surface of the culture tubes. This growth was most marked in the lower half of the tube.

The organism as a rule has not grown in fluid medium as well as in the solid medium.

SUMMARY

We have isolated from the enlarged lymph-nodes of a patient who showed the clinical picture of Hodgkin's disease, and from whom excised glands histologically corresponded to this disease, a pure culture of a pleomorphic, Gram-positive, non-motile, non-acid fast, facultative anaerobic organism, similar to those described by Bunting and Yates.

Cultures of the organism isolated by us have been repeatedly injected into the tissues about the axillae of an adult monkey (*Macacus rhesus*), but up to the time of this report we have obtained no conclusive results.

# THE OCCURRENCE OF MALIGNANT NEOPLASMS IN THE YOUNG

## AS SHOWN BY AN ANALYSIS OF 2,000 CASES OF MALIGNANT NEOPLASMS EXAMINED IN THE PATHOLOGICAL LABORATORY OF THE UNIVERSITY OF MICHIGAN *

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The present study is based on an analysis of 2,000 cases of malignant neoplasm contained in a total of 3,600 cases of neoplasm examined microscopically in the Pathological Laboratory of the University of Michigan during the years 1895-1913. This material is the same as that used in the two papers by Dr. Weller and myself[1] in THE ARCHIVES OF INTERNAL MEDICINE in 1913. As stated there, the material studied is a homogeneous one, derived almost entirely from the state of Michigan, and representing the average population of the state. Further, the material has been utilized not only for the purposes of practical diagnosis, but also in the teaching of pathologic histology, so that the percentage of error in diagnosis can be safely regarded as reduced to a minimum. This point is of especial importance, since the majority of statements concerning the occurrence of malignant neoplasm in the young are to be found in writings of the Virchow period when no exact differentiation of carcinoma and sarcoma was made, and when malignant teratoid tumors were classed as sarcoma or carcinoma or chondroma, etc., as the case might be, according to the preponderance of one or the other of the elements making up the growth. Doubt is, therefore, justly thrown on many of the diagnoses in the reported cases of congenital and early-life tumors. As Williams has mentioned, this is particularly true of the tumors described as "epitheliomata."

The age-period of 1 to 30 years has been chosen, as covering the entire period of development. It is of the greatest importance as far as the broad general neoplasm problem is concerned to establish the relations existing, if any, between the origin of neoplasms and the

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* Read before the Association of American Pathologists and Bacteriologists, Washington, D. C., 1913.
* From the Pathological Laboratory of the University of Michigan, Ann Arbor, Mich.

1. Weller, C. V.: Age Incidence in Carcinoma, THE ARCHIVES INT. MED., 1913, xii, 539. Warthin, A. S.: Heredity with Reference to Carcinoma, etc., ibid., p. 546.

periods of bodily growth. The statement is frequently met in the literature that the development of malignant neoplasms is especially related to the climacterics. It is essential to know if such a relationship exists in the case of the lesser climacterics as well as of the grand. In the consideration of the occurrence of neoplasms in the young the first three decenniums should be taken as covering the periods of dentition, puberty, youth and early sexual maturity, if this question is to be settled. This has not yet been done. The greater number of the reports of malignant neoplasm has been concerned almost wholly with the question of *earliness*. Numerous cases of congenital and early-life neoplasms are recorded in the transactions of pathologic societies with reference to this one point; but little analysis has been made of collected cases from the more important point of view.

Of the 2,000 cases of malignant neoplasms studied, 195, or 9.75 per cent., occurred in the age-period 1 to 30 years. Of these 108 cases, or 55.4 per cent., were in males, and 87, or 44.6 per cent., in females (Table 1). The forms of neoplasms and their distribution through the thirty years is as follows:

AGE INCIDENCE

First Year

Rhabdomyosarcoma of kidney.
Round-cell sarcoma of orbit.
Lymphosarcoma of intestines.
Adenosarcoma of kidney, congenital.
Malignant teratoma of testis.
Angiosarcoma of eye.
Myosarcoma of femur.
Congenital fibrosarcoma of neck.
Round-cell sarcoma of eye.
Spindle-cell sarcoma of jaw.
Adenorhabdomyosarcoma of kidney.

Second Year

Lymphosarcomatosis.
Angiosarcoma of eye.

Third Year

Small spindle-cell sarcoma of radius.
Rhabdomyosarcoma of kidney.
Spindle-cell fibrosarcoma of neck.
Lymphosarcoma of orbit.

Fourth Year

Lymphosarcoma.
Myosarcoma of bladder.
Squamous-celled carcinoma of ear.
Rhabdomyosarcoma of testis.
Small round-cell sarcoma of eye.

Fifth Year

No cases.

Sixth Year

Lymphosarcomatosis.
Round-cell sarcoma of femur.

Seventh Year

Osteochondrosarcoma of femur.

Eighth Year

Spindle-cell sarcoma of orbit.
Large round-cell sarcoma of tibia.

Ninth Year

Large round-cell sarcoma of leg.
Fibrosarcoma of nose.

Tenth Year

Osteosarcoma of foot.
Spindle-cell sarcoma of eyelid.
Round-cell sarcoma of tibia.
Small round-cell sarcoma of orbit.

Eleventh Year

Squamous-celled carcinoma of leg.
Sarcoma of eye.
Lymphosarcoma of intestine.

Twelfth Year

No cases.

Thirteenth Year

No cases.

Fourteenth Year

Spindle-cell sarcoma of ulna.
Squamous-cell carcinoma of mouth.
Carcinoma of stomach.
Osteosarcoma of pelvis.
Adenocarcinoma of umbilicus (congenital).
Basal-celled carcinoma of eye.

AGE INCIDENCE—(CONTINUED)

FIFTEENTH YEAR

Adenocarcinoma of ovary.
Endothelioma of orbit.

SIXTEENTH YEAR

Osteochondrosarcoma of femur.
Alveolar sarcoma of tibia.
Large round-cell sarcoma of uterus.
Medullary carcinoma of ovary.
Large spindle-cell sarcoma of clavicle.
Osteosarcoma of tibia.
Osteochondroma of scapula.
Round-cell sarcoma of femur.

SEVENTEENTH YEAR

Spindle-cell sarcoma of tibia.
Sarcoma of parotid region.
Osteosarcoma of femur.

EIGHTEENTH YEAR

Basal-cell carcinoma of eye.
Malignant teratoma of ovary.
Osteosarcoma of humerus.
Colloid carcinoma of colon.
Round-cell sarcoma of skin of thigh.

NINETEENTH YEAR

Malignant teratoma of ovary.
Small round-cell sarcoma of orbit.
Alveolar round-cell sarcoma of ribs.
Large spindle-cell sarcoma of brain.
Spindle-cell sarcoma of bladder.
Metastatic carcinoma in cervical lymph-nodes.
Squamous-celled carcinoma of cheek.
Melanotic sarcoma of skin.

TWENTIETH YEAR

Colloid carcinoma of intestine.
Spindle-cell sarcoma of parotid.
Malignant syncytioma of uterus.
Malignant teratoma of testis.
Scirrhous carcinoma of stomach.
Fibrosarcoma of jaw.
Small spindle-cell sarcoma of jaw.
Spindle-cell sarcoma of face.
Fibrosarcoma of popliteal space.

TWENTY-FIRST YEAR

Squamous-celled carcinoma of cervix.
Myosarcoma of femur.
Basal-celled carcinoma of conjunctiva.
Lymphosarcomatosis.
Giant-cell sarcoma of ulna.

TWENTY-SECOND YEAR

Squamous-celled carcinoma of skin of abdomen.
Osteosarcoma of pubis.
Giant-cell sarcoma of jaw.
Lymphosarcomatosis.
Squamous-celled carcinoma of eye.
Small round-cell sarcoma of fibula.

TWENTY-THIRD YEAR

Squamous-cell carcinoma of skin.
Carcinoma of breast.
Lymphosarcomatosis .
Basal-cell carcinoma of eye.
Colloid carcinoma of cecum.

TWENTY-FOURTH YEAR

Adenocarcinoma of cecum.
Adenocarcinoma of breast.
Adenocarcinoma of ovary.
Osteochondrosarcoma of pelvis.
Alveolar round-cell sarcoma of neck.
Malignant teratoma of testis.
Fibrosarcoma of nose.
Malignant teratoma of testis.

TWENTY-FIFTH YEAR

Squamous-cell carcinoma of chin.
Medullary carcinoma of breast.
Malignant teratoma of testis.
Osteosarcoma of femur.
Lupus-carcinoma of face.
Lupus-carcinoma of cheek.
Melanotic sarcoma of skin.
Giant-cell sarcoma of jaw.
Myosarcoma of uterus.
Malignant teratoma of testis.

TWENTY-SIXTH YEAR

Sarcoma of skin.
Basal-cell carcinoma of lip.
Round-cell alveolar sarcoma of humerus.
Squamous-celled carcinoma of cervix.
Spindle-cell fibrosarcoma of jaw.
Fibrosarcoma of breast.

TWENTY-SEVENTH YEAR

Adenocarcinoma of uterus.
Carcinoma of antrum.
Spindle-cell sarcoma of breast.
Lymphosarcomatosis.
Medullary squamous celled carcinoma of cervix.
Squamous-celled carcinoma of lip.
Adenocarcinoma of uterus.
Squamous-celled carcinoma of hand.
Malignant teratoma of testis.
Adenocarcinoma of thyroid.
Squamous-celled carcinoma of cervix.

TWENTY-EIGHTH YEAR

Malignant teratoma of testis.
Cystocarcinoma of ovary.
Carcinoma of breast.
Squamous-called carcinoma of lip.
Rodent ulcer of eye.
Scirrhous carcinoma of pylorus.
Malignant teratoma of testis.
Medullary round-cell sarcoma of popliteal space.
Sarcoma of ovary.

AGE INCIDENCE—(CONTINUED)

TWENTY-EIGHTH YEAR—(*Continued*)
Endothelioma of neck.
Malignant teratoma of testis.
Squamous-celled carcinoma of mouth.
Cystadenocarcinoma of breast.
Squamous-celled carcinoma of lip.

TWENTY-NINTH YEAR
Spindle-cell sarcoma of femur.
Carcinoma of parotid.
Cystocarcinoma of ovary.
Endothelioma of orbit.
Squamous-celled carcinoma of penis.
Adenocarcinoma of tube.
Rodent ulcer of inner canthus.
Endothelioma of mouth.
Squamous-celled carcinoma of cervix.
Adenocarcinoma of rectum.
Squamous-celled carcinoma of cervix.
Basal-celled carcinoma of lip.
Medullary carcinoma of breast.
Basal-called carcinoma of nose.
Squamous-celled carcinoma of lip.

THIRTIETH YEAR
Spindle-cell sarcoma of ovary.
Melanotic sarcoma of skin.
Spindle-cell sarcoma of heel.
Medullary carcinoma of cervix.
Endothelioma of chin.
Squamous-cell carcinoma of cervix.
Myosarcoma of cervix.
Lymphosarcoma.
Cystocarcinoma of ovary.
Myosarcoma of uterus.
Squamous-cell carcinoma of penis.
Medullary carcinoma of breast.
Carcinoma of breast.
Medullary carcinoma of cervix.
Medullary carcinoma of breast.
Lymphosarcomatosis.
Squamous-celled carcinoma of cervix.
Medullary carcinoma of cervix.
Adenocarcinoma of rectum.
Adenosarcoma of cervix, congenital.
Malignant syncytioma of uterus.
Lymphosarcomatosis.
Squamous-celled carcinoma of cervix.
Medullary carcinoma of breast.
Scirrhous carcinoma of breast.
Squamous-celled carcinoma of lip.
Cylindroma of parotid.
Squamous-celled carcinoma of leg.
Squamous-celled carcinoma of lip.
Myosarcoma of uterus.

The types of neoplasms were divided as follows: carcinoma 77, or 39.5 per cent.; sarcoma 92, or 47.3 per cent.; malignant teratoma 19, or 9.7 per cent.; malignant syncytioma 2, or 1 per cent.; endothelioma 5, or 2.5 per cent. If the malignant syncytioma and the malignant teratoma be added to the carcinoma, as all of the latter showed atypical epithelial proliferations, even in many cases to the extent of producing epithelial metastases of the nature of carcinoma, and as malignant syncytioma is epithelial in origin, it will be seen that the types of malignant neoplasms in this series are nearly evenly divided between connective-tissue and epithelial growths (Table 1).

TABLE 1.—MALIGNANT NEOPLASMS, 2,000 CASES; IN THE AGE PERIOD 1-30 YEARS, 195 CASES, OR 9.75 PER CENT.

| | Cases | Per Cent. |
|---|---|---|
| Males | 108 | 55.4 |
| Females | 87 | 44.6 |
| Carcinoma | 77 | 39.5 |
| Sarcoma | 92 | 47.3 |
| Malignant Teratoma | 19 | 9.7 |
| Malignant Syncytioma | 2 | 1.0 |
| Endothelioma | 5 | 2.5 |

Of the carcinomas the distribution as to locality was as follows: antrum 1, breast 12, eye and conjunctiva 7, small intestine 1, colon 1, cecum 2, rectum 2, lip 9, lymph-nodes 1, mouth 2, ovary 6, parotid 1, penis 2, tube 1, hand 1, leg 2, ear 1, cheek 2, nose 2, chin 1, elbow 1, stomach 2, pylorus 1, umbilicus 1, cervix of uterus 12, endometrium 2, thyroid 1 (Table 2).

TABLE 2.—Site of Carcinoma

| | | | |
|---|---|---|---|
| Antrum | 1 | Skin, back of hand | 1 |
| Breast | 12 | Leg | 2 |
| Eye, conjunctiva | 7 | Ear | 1 |
| Intestine | 1 | Cheek | 2 |
| Colon | 1 | Nose | 2 |
| Cecum | 2 | Chin | 1 |
| Rectum | 2 | Elbow | 1 |
| Lip | 9 | Stomach | 2 |
| Lymph-Nodes, metastatic | 1 | Pylorus | 1 |
| Mouth | 2 | Umbilicus | 1 |
| Ovary | 6 | Uterus, cervix | 12 |
| Parotid | 1 | Endometrium | 2 |
| Penis | 2 | Thyroid | 1 |
| Tube | 1 | | |

The distribution of the sarcomas with respect to site was: bladder 2, jaw 7, femur 10, tibia 6, fibula 1, heel 1, foot 1, pelvis 3, humerus 2, ulna 2, radius 1, rib 1, scapula 1, clavicle 1, brain 1, breast 2, eye 5, eyelid 1, orbit 5, face 1, intestine 2, lymph-nodes 10, muscle of abdomen 2, neck 4, nose 2, ovary 2, parotid 3, popliteal space 2, skin 5, body of uterus 4, cervix 2 (Table 3).

TABLE 3.—Site of Sarcoma

| | | | |
|---|---|---|---|
| Bladder | 2 | Breast | 2 |
| Bone and Periosteum— | | Eye | 5 |
| Jaw | 7 | Eyelid | 1 |
| Femur | 10 | Orbit | 5 |
| Tibia | 6 | Face | 1 |
| Fibula | 1 | Intestine | 2 |
| Heel | 1 | Lymph-nodes | 10 |
| Foot | 1 | Muscle, abdominal | 2 |
| Pelvis | 3 | Neck | 4 |
| Humerus | 2 | Nose | 2 |
| Ulna | 2 | Ovary | 2 |
| Radius | 1 | Parotid | 3 |
| Rib | 1 | Popliteal space | 2 |
| Scapula | 1 | Skin | 5 |
| Clavicle | 1 | Uterus | 4 |
| Brain | 1 | Cervix | 2 |

The distribution of the malignant teratomas was: cervix of uterus 1, kidney 4, ovary 2, testis 12. The endotheliomas were distributed as follows: chin 1, orbit 2, mouth 1, neck 1 (Table 4).

TABLE 4

| Site of Malignant Teratoma | | Site of Endothelioma | |
|---|---|---|---|
| Cervix uteri | 1 | Chin | 1 |
| Kidney | 4 | Orbit | 2 |
| Ovary | 2 | Mouth | 1 |
| Testis | 12 | Neck | 1 |

The chart shows graphically the age distribution. It will be seen by this that the curve is high during the first year, falls then to rise at puberty, after which there is a steady rise up to the limit of the

period, age 30. The large number in the first year emphasizes the importance of the *congenital factor,* and this influence is felt in the case of the majority of the tumors found up to the period of sexual ripeness, even when no actual evidences of a teratoid nature could be demonstrated in the tumor. Sarcoma predominates in the period of infancy and childhood; but the *parallel increase of both sarcoma and carcinoma with age is noteworthy and in contradiction to the usual belief that sarcoma is essentially a disease of childhood.* The cases of malignant teratoma fall in the periods of early childhood and developing sexual life. Particularly is this the case with the malignant tera-

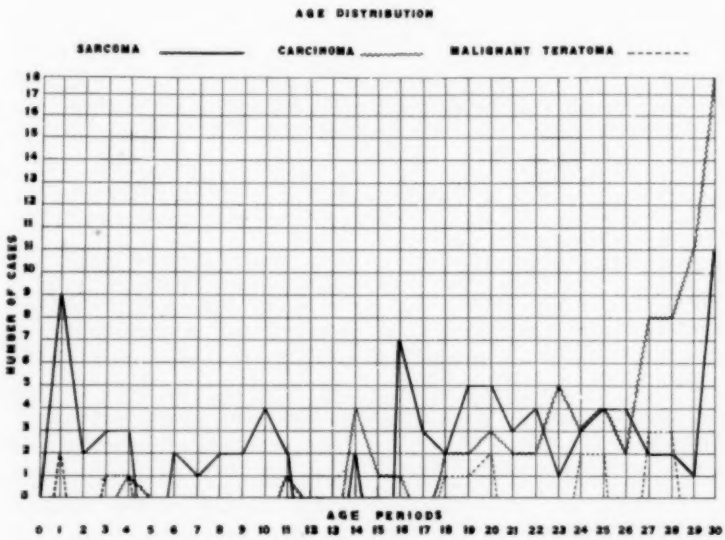


Chart showing age distribution of sarcoma, carcinoma and malignant teratoma.

tomas of testis and ovary, the majority of these cases developing after puberty and in the early period of sexual maturity. No especial relationship to the period of dentition is shown. We had no cases in the fifth year, and none in the twelfth and thirteenth years. The number of cases in the years 6 to 11 is too small to show any conclusion. The main facts shown by the chart are the relatively high number of congenital tumors, the gradual increase in incidence of tumors from the age of adolescence into mature life, the relation of malignant teratomas to puberty and the parallel incidence of carcinoma and sarcoma.

2. Williams: The Natural History of Cancer, 1908.

The best analysis of congenital and early-life neoplasms is that by Williams.[2] The analysis of our 195 cases of malignant neoplasms of this period differs in a number of ways from his. His analysis is based on Duzan's analysis of 182 malignant tumors of early infancy, Picot's analysis of 424 cases, his own of 56 consecutive cases, Poinsot's 25 cases of bone sarcoma in infancy, Gross' study of sarcoma of the long bones (51 cases) and 41 cases tabulated by Butlin and Colby. Williams points out the relative frequency of retinal glioma and renal tumors as congenital. No retinal glioma occurs in my series, and only four cases of congenital neoplasm of the kidney. Likewise the tendency to sarcoma observed by him on the part of the scapula is not shown in our series, only one case being includetl.

Williams concludes that prenatal life, infancy and childhood are wholly exempt from malignant epithelial tumors. He rejects many of the reported cases as untrustworthy. He gives the earliest age at which carcinoma has been positively demonstrated as 11 years (columnar-celled cancer of rectum). He himself saw a case of rodent ulcer at 14 years. He asserts that under the age of puberty cancer is practically unknown. In our list we find an undoubted and typical squamous-celled horny cancer of the lobe of the ear at 4 years (no evidence of teratoid origin could be found); at 11 years a typical horny squamous-celled carcinoma of the skin of the leg; in the fourteenth year an adenocarcinoma of the umbilicus (teratoid in origin), a typical squamous-celled carcinoma of the mouth, a basal-celled carcinoma of the eye and an adenocarcinoma of the stomach. Our list then gives undoubted examples of earlier carcinoma than those recorded by Williams. He also reports the earliest examples of cancer of the breast as occurring at 20.5 and 21 years. Our list gives two operated on in the twenty-third year, the tumors having begun to develop before the twentieth year.

My conclusions in the main agree with those of Williams. Malignant tumors are relatively rare before puberty, but there is a steady ascending line of occurrence from childhood onward to middle life, *both sarcoma and carcinoma showing a parallelism of occurrence.* This fact is also strikingly brought out in the study of sarcoma by Dr. Weller. Our cases in a large measure also support Williams' view of the *congenital or teratoid origin of malignant neoplasms in infancy and early life.*

*The tendency to malignant tumors is relatively slight before sexual maturity is reached, increases during the period of sexual maturity, up to the age-period of 58 to 62. With the decline of the organism the tendency to malignant neoplasm decreases.*

# EXPERIMENTAL DIABETES INSIPIDUS IN DOGS*

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It is a well substantiated clinical observation that injuries involving the base of the cranium frequently are followed by polyuria which may or may not be accompanied by glycosuria. It is also just as well substantiated that growths which involve the base of the brain, especially the region of the third ventricle, may likewise give rise to polyuria which may be accompanied by hyperglycemia sufficient to produce glycosuria. These conditions when arising from fractures of the base of the skull generally are of short duration, but when they result from growths in the region above mentioned, may be more or less persistent.

The first experimental evidence that injuries to the base of the brain (in the region of the fourth ventricle) gave rise (under certain conditions of the animal operated on) to polyuria accompanied by hyperglycemia and glycosuria was furnished by the classic experiment of Bernard known as the Bernard piqûre. Further, it has been observed that certain piqûres in the base of the brain may give rise to polyuria alone without glycosuria. As Bernard failed to induce any glycosuria by his piqûre after section of the splanchnic nerves, or in fasting animals, our present-day interpretation of his results would be that he was dealing with an epinephrin glycosuria. However that may be, Cushing[1] and his coworkers have thrown some doubt on this interpretation, as well as on the assumption of the existence of a diabetic center in the floor of the fourth ventricle, and have brought forward much experimental evidence to show that the glycosuria induced by the Bernard piqûre is in all probability of hypophyseal origin. The same evidence is applicable to the polyuria induced by piqûres further forward in the brain.[2]

All workers in this field have noted that the injection of extracts of the posterior lobe cause a rise in the blood-pressure generally accompanied by a transient (more lasting than the increase in blood-pressure) polyuria. Like observations[3] on the volume of the kidneys

* Submitted for publication Aug. 31, 1914.

1. Cushing, H.: Bull. Johns Hopkins Hosp., 1913, xxiv, 40.

2. Cushing, by stimulating certain nerves of the autonomic system (Langley) which make connection with the pituitary body, and which he claims controls its secretion, was able to bring about these conditions (polyuria and glycosuria), which he explains in terms of hypersecretion of the nervous part of the gland.

3. King and Stoland: Am. Jour. Physiol., 1913, xxxii, 405.

show the following general results: Immediately following the beginning of the rise in blood-pressure there is a contraction of the kidneys, during which time the secretion of the urine is almost suspended. Following this initial contraction and anuria the kidneys begin to dilate and often attain a degree of dilatation much above the normal. Accompanying and during the time the kidneys remain dilated (about thirty minutes from a single dose) there is a marked polyuria, sometimes glycosuria. The intensity of the diuresis induced is always proportional to the degree of kidney dilatation and only lasts as long as the kidneys remain dilated. This is paramount to saying that the diuretic effect of posterior lobe extracts is due to a local dilatation of the kidney vessels, thus allowing a greater quantity of blood to pass through the kidneys per unit of time than under normal conditions, just as the diuresis induced by caffein or strophanthus can be accounted for in terms of kidney volume.

Nearly all the investigations so far on the function of the pituitary body have brought out the fact that certain injuries of the posterior lobe or of the stalk, such as partial removal of the lobe or irritation of the stalk, frequently are followed by polyuria; sometimes, though less frequently, by glycosuria, generally transient but sometimes persisting for weeks, in which case the polyuria is accompanied by a great laying-on of fat.[4] In two young female dogs I was able to induce a permanent polyuria (a veritable diabetes insipidus) accompanied by a rapid accumulation of fat by operating on the base of the brain as follows:

A hole about 5 mm. in diameter was drilled with an ordinary dental burr, driven by a dental engine, from the roof of the mouth up through the sphenoid bone into the floor of the sella turcica. The opening removed the posterior rim of the pituitary fossa and extended back about 5 mm. This exposed the posterior surface of the posterior lobe and the region just posterior to it. The hole in the sphenoid bone was then plugged with a gutta-percha compound such as is used for temporary fillings in teeth. The plug was placed so that it would impinge somewhat on the posterior lobe and its stalk and extend up into the third ventricle.

Both animals recovered from the effects of the operation, and the wounds healed without infection. No nasal trouble followed. No sugar was found in the urine secreted during the time of the operation, nor at any subsequent time. Only a small amount of urine was passed (about 150 c.c.) during the forty-eight hours following the operations. After the second day polyuria began to develop. One of these animals in particular (weight 6 kg.) after the fourth day required from 5 to 6 liters of water every twenty-four hours to make it comfortable and passed from 5 to 6.5 liters of urine per day for nine weeks, and in the same time gained three kilos in weight. While the animal was not eating, drinking and urinating, it simply slept and grew fat. With an abundance of water (6 to 7 liters) the amount of urine voided per twenty-four hours was about equal to the amount of water imbibed; but if water was given more sparingly (4 liters) the tendency was to pass more urine than the

4. Cushing, H.: Boston Med. and Surg. Jour., 1913, clxviii, 901.

water imbibed. If given an ordinary amount of water (300 to 500 c.c.) about 600 to 800 c.c. urine would be passed, and the animal would show signs of distress. The dog enjoyed a good appetite throughout, eating well of table scraps, etc. The sugar tolerance was high; in fact it was impossible to induce an alimentary glycosuria, the animal being able to ingest 15 gm. of glucose per kilo body weight in six hours without showing any signs of glycosuria.

The urine passed was simply a dilute normal urine (specific gravity 1.001-1.002). The amount of nitrogen eliminated when computed in terms of the food eaten and a normal amount of urine (250 to 300 c.c.) was about normal. Also the relative amounts of the different nitrogen constituents of the urine were normal. In fact, there was no change in the nitrogen metabolism. The inorganic salts were approximately normal. With a given amount of water, sufficient to maintain physical comfort (6 liters), the concentration of the urine remained about constant (specific gravity 1.001-1.002). With less water the urine tended to become more concentrated. Ordinary quantities of water, less than 2 liters per day, caused great physical discomfort.

During the animal's life after the operation (nine weeks) it gained 3 kilos (50 per cent.) which seemed to be almost wholly represented by an accumulation of fat. While it was found impossible to induce an alimentary glycosuria, the animal responded promptly to phlorizin with a copious output of sugar.[5]

The dog's temperature remained low throughout, not more than 37 C., and the state of lethargy in which it remained indicated profound hypopituitarism as defined by Cushing.

*Necropsy.*—The hole in the sphenoid bone included about 1 mm. of the posterior part of the pituitary fossa and extended posteriorly about 4 mm. The plug of temporary dental stopping had healed into the bone and protruded up along the posterior surface of the gland for about 4 mm. It had impinged somewhat on the nervous portion of the gland and the point of the plug was in close proximity to the stalk at its entrance into the floor of the third ventricle. The plug had exerted considerable pressure on the posterior lobe and the stalk, but not sufficient to destroy the gland, nor materially to change its structure. The anterior lobe was apparently normal and the base of the brain was perfectly clean.

Outside of the cranium the principal pathological finding was an excessive accumulation of fat, not only in the regions of the body where fat normally deposits, but all the tissues were infiltrated with fat. It was noticed before death that the heart was not working well; and when opened deposits of fat were found on the valves and in the interstices of the papillary muscles sufficient to occupy a large part of the space in the heart's cavities. In fact, all the spaces and interstices in the muscles of the body everywhere were distended with fat. The muscle cells themselves were not well nourished. All the glandular organs were infiltrated with fat, and the liver especially showed numerous necrotic areas. The kidneys were small and the cells contained an excess of fat. The glomeruli were full of fat. No marked inflammatory changes had occurred in the tubular epithelium, although the cells did not stain well.

Cushing and his coadjutors as well as nearly all other workers in this field have described the occurrence of intense polyurias, generally of short duration, following the partial removal of the nervous lobe of

5. An attempt was made to inhibit the formation of fat in these dogs by keeping them under the influence of phlorizin, but abscesses tended to form at the site of the injections, so this had to be discontinued. In fact, both animals had to be sacrificed on account of abscess formation due to hypodermic medication.

the gland and also after insult to the stalk. Some of the animals described exhibited a moderate polyuria lasting for months, accompanied with an abnormal accumulation of fat.

There is sufficient evidence now at hand to establish beyond doubt that certain manipulations of the posterior lobe, such as partial removal or injuries to the stalk, will induce a temporary polyuria and that continuous irritation of a small degree, such as slight pressure, may bring on a permanent polyuria (diabetes insipidus). Cushing[4] evidently brought on this condition in one patient by a sellar decompression operation, in which only a small fragment of anterior lobe tissue was removed. In dogs, experimental diabetes insipidus is accompanied by marked symptoms of hypopituitarism and an accumulation of fat. From the clinical reports at hand, this rapid accumulation is not so apparent, although nearly all the other signs of hypopituitarism have been described.[6]

To form a true picture from the experimental data at hand of the mechanism which operates to induce polyuria and polydipsia is quite impossible. One might imagine that by certain manipulations of the gland, such as has been described and which are followed by polyuria and polydipsia, the posterior lobe can be thrown into a state of hypersecretion; and that the excess of the secretion acts in the same manner as the injection of posterior lobe extract. This would give rise to a general vasomotor contraction of short duration followed by a local vasomotor dilatation of the kidney vessels of a more lasting character. Such a change in the relative blood-supply between the body as a whole and the kidneys always gives rise to diuresis. In fact, all chemical compounds possessed of diuretic properties act on the vascular system in this way (caffein; strophanthus; pituitrin). Should one attempt to explain the polyuria and polydipsia arising from hypophyseal derangements, such as has been described in terms of the diuresis caused by the injection of extracts of the posterior lobe, the following explanation (more or less transcendental) seems to satisfy at least a number of the observed facts:

To conform with this generally accepted interpretation of the action of extracts of the posterior lobe in causing diuresis, the kidneys will have to be looked on as dialyzing membranes and the urine as a dialysate. This conception makes the dialyzing membranes (kidneys) permeable to all the normal constituents of the urine, and the rate of dialysis to depend on the concentration in the blood of the constituents of the urine, and the amount of the solution of these substances (blood) which comes in contact with the dialyzing membranes per unit

6. Marie and Boutier: Rev. neurol., 1913, xxv, 555.

of time. Such a mechanism as this would tend to deplete the body of water as well as of the effete materials given off from the tissues into the blood, the solution of which in water constitutes the urine.[7]

Ordinarily, the circulation through the kidneys is about sufficient to permit of the passage of about 1,500 c.c. of water with its contained substances (urine) per day. To maintain life and comfort this water must be replenished from the outside, which is accomplished by absorption from the stomach and intestines. When a certain amount of water has passed from the body a disagreeable sensation begins to manifest itself in the mouth and the pharynx, which is relieved by the imbibition of water. When water passes rapidly through the kidneys or is given off from the surface of the body in large amounts, this sensation occurs at frequent intervals, and if the excretion of water is very rapid, it may be almost continuous (polydipsia). This is analogous to the sensation referable to the stomach called hunger. When the tissues begin to suffer from lack of food, some influence begins to exert itself on the stomach, causing contractions which give rise to painful sensations called hunger pains, and which are relieved by the taking of food. In diabetes mellitus the body cannot utilize one of the chief food substances (carbohydrates) and in extreme cases hunger pains are almost constant.[8] Likewise when the kidneys permit the passage of an excessive amount of water, polydipsia results.

As already stated, diuresis results whenever an excess of blood is permitted to pass through the kidneys in a given time, as compared with the blood passing through the remainder of the body in the same time. This deviation from the normal relation of the kidney blood-supply to the tissues in general would tend to draw off the water from the body to an extent sufficient to cause polydipsia. Should the kidneys attain to a great degree of dilatation, such as often occurs after the injection of pituitrin, the elimination of water might be sufficiently rapid to cause a constant state of polydipsia, and if one were allowed to imagine a condition of prolonged dilatation of the kidneys to result, a state of polyuria and polydipsia comparable to the dog described might well result.

While this explanation satisfies quite well most of the conditions present in the polyurias and polydipsias of hypophyseal origin, both clinical and experimental, it can hardly be said that there is sufficient

7. Water and the inorganic salts are essential to life and must therefore be considered as foods. If the effete materials which result from cell metabolism appear in the urine as dialysates they must first be held in a state of solution. So it would appear that one of the functions of water in the animal economy is that of a solvent for these effete substances, and that its excretion is necessary to accomplish their excretion.

8. Luckhardt: Am. Jour. Physiol., 1914, xxxiii, 313.

experimental evidence at hand to support it unconditionally, although experimental evidence in its support is not wholly wanting. We can hardly say, however, that we have sufficient evidence at hand to prove that operations on the pituitary body such as are followed by polyuria and polydipsia, or that such pathological lesions as give rise to polyuria, cause a hypersecretion of the posterior lobe of the gland sufficient to keep the kidneys in a dilated state for weeks or months. Neither have we sufficient evidence to claim that a constant hypersecretion of the posterior lobe will keep the kidneys in a state of dilatation, nor do we know that repeated doses of posterior lobe extract over a long time will accomplish any such results. In fact, Farini,[9] P. Bioch,[10] R. Balnet[11] and von den Velden[12] claim that in human subjects the injection of posterior lobe extracts exerts no diuretic influence at all, and that when administered to patients suffering from diabetes insipidus they reduce the amount of urine passed to or below normal. In normal dogs injections of posterior lobe extract twice daily failed to modify the amount of urine passed over a period of seven days.

Direct stimulation of the posterior lobe or of the nerves going to it generally cause a contraction of the kidneys which may last for a few minutes only (ten to fifteen) to be followed by a dilatation accompanied by diuresis, or the initial contraction may last for hours (twenty-four to forty-eight) eventually to be followed by polyuria, which argues for a dilatation of the kidneys. Frequently, while drilling through the sphenoid bone in pituitary operations, an oncometer record of the kidney volume being taken at the same time, I have observed a contraction of the kidney which sometimes continued throughout the operation with almost complete suppression of the urine. In fact, anuria may continue for forty-eight hours after the operation. Under like conditions I have noted an initial contraction of the kidney to be followed in from ten to fifteen minutes by dilatation and diuresis, and at other times and under identical conditions a dilatation of the kidneys from the first accompanied by diuresis.

While these observations seem contradictory, nevertheless, when scrutinized, their lack of agreement is more apparent than real. The experimental evidence seems to bring out quite clearly that continued stimulation (irritation) not of sufficient intensity to destroy the integrity of the gland, may give rise to a state of polyuria and polydipsia of long duration, lasting at least as long as the stimulation lasts, provided, however, the integrity of the gland is preserved. Operations,

9. Farini: Riforma med., 1913.
10. Bioch, P.: München. med. Wchnschr., 1914, lxi, 217.
11. Balnet, R.: Berl. klin. Wchnschr., 1913, l, 2379.
12. von den Velden: Berl. klin. Wchnschr., 1913, l, 2085.

such as direct stimulation of the gland for a short time only, or partial or total removal of the gland, induce a diuresis of short duration, and if we admit that the diuresis results from a setting free of an excess of the gland's secretion, as long as the effects of the excess secretion lasts. As to whether in these cases of experimental diabetes insipidus the volume of the kidneys in increased, we have no direct evidence. In fact, we have no method at hand to demonstrate whether or not such a condition exists.[13]

As for the explanation of the excessive formation of fat under these conditions, none is at hand. Carbohydrates seem to be rapidly converted into fat, which may account for the high sugar tolerance of the animals. At any rate it seems that the food value of the carbohydrates is reduced very much under the conditions imposed, and that the real condition of the body at the end is that of emaciation. In diabetes mellitus the body cannot use the sugars, so in experimental diabetes insipidus of pituitary origin in dogs the body does not seem to be able to utilize either the sugars or the fats to their full extent.

13. The theory that in diabetes insipidus some substance is liberated in the body that renders the kidney epithelium permeable only to dilute solutions, could not be made to hold in these dogs. The kidneys here were permeable to a urine, having a specific gravity of 1.010, as shown by the imbibition of salt and limiting the water supply.

# THE USE OF STRYCHNIN AND CAFFEIN AS CARDIOVASCULAR STIMULANTS IN THE ACUTE INFECTIOUS DISEASES *

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For decades many physicians have relied, with unquestioning faith, on strychnin in the treatment of certain grave symptoms occurring in pneumonia, typhoid fever and other acute infectious diseases. Within the last twenty-five years some physicians have preferred to use caffein (and other drugs) when confronted with these symptoms. The symptoms referred to are those which are thought to signify an approaching or an already existing failure of the circulation, and strychnin and caffein are often believed to be powerful and rapid stimulants for the cardiovascular apparatus in such a state.

It is the object of this communication to discuss briefly the question whether failure of either the heart or the vasomotor apparatus is the chief cause of death in the infectious diseases; and to examine, with the aid of new data, the pharmacological and clinical evidence for and against the use of strychnin and caffein as cardiovascular stimulants in infections.

## THE ALLEGED FAILURE OF THE CIRCULATION

A normal flow of blood is brought about chiefly by two factors — a heart which pumps a sufficient amount of blood, and a peripheral resistance maintained by the partial contraction of the small arteries. The tone of the arteries depends, in its turn, on the normal activity of the vasomotor nervous system.

*The Heart.*—It is one of the axioms of clinical medicine that the heart-muscle may be so seriously damaged in the acute infections as to be an important source of death. The symptoms which are alleged to signify approaching heart failure of this type are rapid pulse, irregular pulse, dyspnea, cyanosis, increased area of cardiac dulness and weak heart sounds. Space does not permit a discussion of each one of these symptoms. Occasion will, however, be taken to point out

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that at least the cyanosis[1] and dyspnea[2] may not be of cardiac origin; that an increased area of dulness is a rare finding in this connection, and that most clinicians have great difficulty in judging of the relative strength of heart-sounds.

Nor is the experimental evidence more satisfying. In 1899, Romberg, Pässler, Bruhns and Müller[3] investigated, by means of experiments on animals, the cause for the cardiovascular disturbances of the acute infectious diseases. They produced a fatal pneumococcus septicemia, fatal diphtheria, and a fatal pyocyaneous infection in rabbits. Even when the animals were near death, upward stroking of the abdomen still caused a moderate rise in blood-pressure, and clamping the abdominal aorta was still followed by a sharp rise in pressure. Romberg and his coworkers argued that if the heart-muscle were exhausted, it would yield before an increasing load, and so the two above-mentioned maneuvers would not result in a rise of blood-pressure. Stejskal[4] disagreed with their conclusions because he was able to show that abdominal massage could increase arterial pressure even in a dead animal. Gottlieb[5] also felt that Romberg's data did not justify his conclusions. MacCallum[6] has recently reinvestigated the state of the heart in fatal diphtheria intoxication. He says in one place:

> There is no comparison between the perfused normal heart and the diphtheria hearts, all of which are feeble and apt to go into fibrillation. Nevertheless, although one may receive the impression that the hearts from the poisoned animals are rather weak and apt to be irregular, it is clear that they continue to beat for several hours after they have shown every sign of failure in the body of the dying animal, if only the pressure of nutritive fluid be maintained in the coronary arteries. Indeed, the animal may be allowed to die, and an hour after its death the heart can be revived and will beat for a long time. All of this seems to show fairly well that the death which occurs in the height of an attack of diphtheria is not exclusively the result of direct injury to the heart, although that may play some part in the process.

Such are the data concerning the state of the myocardium in the acute infectious diseases. It is difficult to draw conclusions from them. Certain symptoms, such as rapid and irregular pulse, and dyspnea

1. Peabody, F. W.: The Oxygen Content of the Blood in Lobar Pneumonia, Jour. Exper. Med., 1913, xviii, 7.

2. Barcroft, J.: The Respiratory Function of the Blood, 1914, Camb. Univ. Press.

3. Romberg, Pässler, Bruhns and Müller: Exp. Untersuchungen über die allg. Path. der Kreislaufstörung bei acuten Infectionskrankheiten, Arch. f. klin. Med., 1899, lxiv, 652.

4. Stejskal, K. R.: Kritisch. Exp. Untersuchungen über den Herztod in Folge von Diphtherietoxin, Ztschr. f. klin. Med., 1902, xliv, 367.

5. Gottlieb: Med. Klin., 1905, p. 25.

6. MacCallum, W. G.: The Mechanism of the Circulatory Failure in Diphtheria, Am. Jour. Med. Sc., 1914, cxlvii, 37.

occurring in the course of acute infections, may logically be offered as evidence that the heart is seriously threatened. On the other hand, the data collected from experiments on animals fail to show that the heart-muscle is exhausted by these diseases. But these latter data are not entirely satisfactory, because, being negative, they are open to the criticism that a new method of attack might bring forth positive evidence of serious myocardial injury.

*The Peripheral Resistance.*—Having proved to their satisfaction that the heart could not be directly responsible for the disturbances in question, Romberg and his associates presented further data which they interpreted to mean that the vasomotor apparatus was paralyzed in these diseases. Clinicians accepted this hypothesis and quickly began to direct their therapeutic efforts against this new agent of death. A new reason now existed for the use of strychnin and caffein in these diseases, because both of these drugs were said to be powerful vasomotor stimulants.

But it has been recently shown that the vasomotor apparatus does not fail in pneumonia or diphtheria. Porter and Pratt[7] and Porter and Newburgh[8] have found the vasomotor reflexes normal in all stages of diphtheria and pneumonia; and Newburgh and Minot[9] have shown that the blood-pressure is usually not low in fatal pneumonia. If the vasomotor apparatus is normal in pneumonia and diphtheria, it may be assumed that it also maintains its integrity in other acute infections. Such an assumption is, at least, justified until evidence against it is presented.

To sum up this portion of the discussion, it may be stated (1) that since there is no evidence that the vasomotor apparatus is injured in the acute infectious diseases, it is not logical to direct treatment chiefly toward this apparatus; (2) that the hypothesis that the heart may be fatally injured in the acute infections, although far from proved, is still tenable in the present state of our knowledge, and that it is consequently logical to attempt to assist it when it seems to be failing.

We turn now to strychnin and caffein, two of the drugs so frequently used as cardiovascular stimulants.

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7. Porter and Pratt: The State of the Vasomotor Center in Diphtheria Intoxication, Am. Jour. Physiol., 1914, xxxiii, 431.

8. Porter and Newburgh: The State of the Vasomotor Apparatus in Pneumonia, Am. Jour. Physiol., 1914, xxxv, No. 1.

9. Newburgh and Minot: The Blood-Pressure in Pneumonia, THE ARCHIVES INT. MED., 1914, xiv, 48.

STRYCHNIN

Strychnin is used in the treatment of the acute infectious diseases because it is believed that it raises the blood-pressure, slows the pulse and increases the force of the heart. But a search through the literature shows that there is not a single piece of experimental evidence to warrant such a belief.

In fact, Mayer[10] in 1871 and Denys[11] in 1885 showed that strychnin in medicinal doses did not change the blood-pressure or the rate of the heart beat. They studied the effect of increasing doses of strychnin on the pressure in the carotid artery of animals and found that the pressure did not rise until the dose was sufficiently great to cause convulsive twitchings, which began simultaneously with the upward movement of the pressure curve. It is important to note that Mayer and Denys worked with non-curarized animals, for results obtained in animals which have been given enough curare to abolish the reflex muscular contractions leave no means of determining whether an amount of strychnin sufficient to raise blood-pressure is not also great enough to cause convulsions. One is not justified in concluding that because strychnin, if given in sufficiently great doses, has a pressor effect in curarized animals, it will have the same effect when given to human beings in medicinal doses.

When the use of strychnin in myocardial disturbances is impartially investigated, it is found that the evidence all points in one direction. Pharmacologists[12] have shown that strychnin is without effect on the heart until given in doses so large that death from convulsions invariably ensues. Clinicians[13] have demonstrated that strychnin is of no benefit in the treatment of acute or chronic heart disease.

There are two clinical studies of the effect of strychnin in the acute infectious diseases.

Cook and Briggs[14] in 1903, recorded the effects of stimulants in hypotensive states. In summing up their results with strychnin they said in part:

> Strychnin produces a rise in blood-pressure varying in average cases from one to four hours, and accompanied, in the majority of patients carefully observed, by an improvement in the patient's general condition. When the

10. Mayer, S.: Ber. d. kais. Acad. d. Wissensch., 1871, lxiv, 657.
11. Denys, J.: Arch. f. exper. Path. u. Pharmakol., 1885, xx, 306.
12. Ingersheimer, J.: Ueber die Wirkung des Strychnines auf das Kalt-und Warmblüterherz., Arch. f. Exper. Path. u. Pharmakol., 1905, liv, 73.
13. Parkinson and Rowlands: Strychnin in Heart Failure, Quart. Jour. Med. 1913, vii, 42. Newburgh, L. H.: On the Use of Strychnin in Broken Cardiac Compensation, Am. Jour. Med. Sc., 1915, cxlix.
14. Cook and Briggs: Clinical Observations on Blood-Pressure, Johns Hopkins Hosp. Rep., 1903, xi, 451.

routine administration of strychnin has continued for from eight to twelve doses, individual doses often fail to produce any marked immediate rise in the systolic blood-pressure; but in such cases, if one or two doses are omitted and the pressure is carefully followed in the interval, it will be seen that there is a progressive fall in blood-pressure in the absence of stimulation, the previous level being again restored when the drug is renewed. On the whole, strychnin is by far the most satisfying cardiovascular stimulant for long continued routine administration.

Such results should have been easy of confirmation. Cabot[15] undertook this task the next year. His observations include five thousand measurements in thirty-one cases of typhoid fever, four of pneumonia and fifteen others. The total result was negative. He was unable to convince himself that strychnin exerts any influence on the blood-pressure in febrile cases. Cabot's experience was such as an acquaintance with the pharmacology of the drug would lead one to expect. I shall present data which are in entire accord with those collected by Cabot.

This review of the studies of the action of strychnin must make it evident that there is no reason for prescribing it as an aid to a failing myocardium. Cook and Briggs' enthusiastic recommendation of it as a powerful pressor agent is not supported by laboratory data and is directly refuted by Cabot's investigations.

### CAFFEIN

Caffein has also been credited with the ability to stimulate the vasomotor mechanism and to increase the effectiveness of the heart beat. When we examine the evidence on which the use of caffein as a vasomotor stimulant ought to be based, we find that there has been constant disagreement among experimenters. Some have asserted that caffein had a powerful pressor effect through its action on the vasomotor center. Others have stoutly denied this. In a recent review of the whole subject the reasons for the confusion have been pointed out and new data have been added which finally place the action of caffein on the cardiovascular apparatus on a firm basis.[16] This work presents the following summary:

The circulatory effects of moderate doses of caffein consist in vasodilatation combined with sufficient cardiac stimulation to maintain or even increase blood-pressure. Both of these activities favor blood flow. The oncometer shows conclusively that the moderate rise of pressure which is observed in anesthetized animals is, in most cases, exclusively cardiac.

15. Cabot, R. C.: Measurements of Blood-Pressure in Fevers, Before, During and After Administration of Strychnin, Trans. Assn. Am. Phys., 1904, xix, 22.

16. Sollmann and Pilcher: The Actions of Caffein on the Mammalian Circulation, Jour. Pharm. and Exper. Therap., 1911, iii, 19.

Caffein then does not stimulate the vasomotor center, but it does augment the circulation in the way described above, at least in experimental animals whose hearts were presumably normal. Whether it has a similar beneficial effect in diseased hearts remains still to be demonstrated.

### NEW OBSERVATIONS OF THE EFFECTS OF STRYCHNIN AND CAFFEIN

*Method.*—We have collected clinical data in regard to the question whether strychnin and caffein are valuable cardiovascular stimulants. The observations were made on patients in all stages of acute infectious diseases and in a few others who exhibited very low blood-pressure.

The effect of single doses of varying size, or a number of doses given within a short period of time, on the systolic, diastolic, and pulse-pressure, on the rate and character of the pulse, and the respiratory rate, and on the general condition, was studied. It has been our invariable procedure to measure the blood-pressure, to count the pulse, and usually the respiration, several times immediately before giving the drug and every few minutes thereafter, until it was certain that either no effect had been produced or that the effect had come to an end. Repeatedly, such measurements were made two or more times daily throughout the period of illness, so that the drug period could be compared over several days with the period preceding it and the period following it.

*Limitations of the Method.*—When judging of the effect of any therapeutic procedure on the blood-pressure, one must thoroughly realize that moderate and even large variations result from slight external stimuli of all sorts. For example, Zabel,[17] who investigated the causes for sudden changes in pressure, found that a superficial pin prick caused a systolic rise of 72 mm. in one individual and that the average rise following this stimulus lay between 11 and 30 mm. External cold always caused a rise. Even the application of the unheated cuff of the blood-pressure apparatus caused a variation of 23 mm. in one case. The sight of food produced a considerable rise, drowsiness a fall. Psychic stimuli caused variations of from 30 to 46 mm. He found a normal daily afternoon rise. Even if the measurements were carried out under ideal conditions, that is, by the same observer on the same subject, in bed, in a room by himself, on a uniform diet, the temperature of the room being constant, and the subject being in good health and not exposed to any unpleasant sensa-

17. Zabel: Plötzliche Blutdruckschwaukungen und ihre Ursachen, München. med. Wchnschr., 1910, lvii, 2278.

TABLE 1.—Showing the Effect of Strychnin in Single and Repeated Doses on Systolic and Diastolic Blood-Pressure, Pulse and Respiration

| Type | Case No. | Treatment | Systolic Pressure. Duration of Rise or Fall * | Diastolic Pressure. Duration of Rise or Fall * | Pulse. Duration of Increase or Decrease in Rate * | Respiration. Duration of Increase or Decrease in Rate * |
|---|---|---|---|---|---|---|
| Mild typhoid | 10 | Strychnin, gr. 1/20 subcutaneously. | ↑ 10-13 for thirty minutes. | 0 | 0 | Not noted |
| Same case | 10 | Strychnin, gr. 1/10 subcutaneously. | ↑ 10 for four minutes | ↓ 10 for forty minutes | 0 | Not noted |
| Mild typhoid† | 11 | Strychnin, gr. 1/10 subcutaneously. | ↑ 12-16 for thirty minutes. | ↑ 30 for fifteen minutes | 0 | Not noted |
| Mild lobar pneumonia | 12 | Strychnin, gr. 1/10 subcutaneously at 10:15 and 11:15 a. m. | ↓ 10 for two hours. | ↓ 15 for one and one-half hours. | ↓ 10 for remainder of day. | ↓ 5 for two hours. |
| Same case | 12 | Physical examination next morning. | ↑ 20-30 for one hour + | ↑ 25 for ? | Not noted | Not noted |
| Mild lobar pneumonia | 13 | Strychnin, gr. 1/10 subcutaneously at 12 and 1:30 p. m. | 0 | 0 | ↑ 5-10 for three hours | 0 |
| Mild lobar pneumonia | 14 | Strychnin, gr. 1/10 subcutaneously at 4 and 5:15 p. m. | ↑ 8 for one hour. | 0 | 0 | ↑ 4 for two hours |
| Fatal lobar pneumonia | 15 | Strychnin, gr. 1/10 subcutaneously at 9:20 and 10:05 a. m. | ↑ 5-10 for two hours. | 0 | 0 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| Severe lobar pneumonia | 16 | Strychnin, gr. 1/20 subcutaneously. | 0 | ↑ 5 for one-half hour | 0 | Not noted |
| Same case | 16 | Strychnin, gr. 1/10 subcutaneously. | 0 | 0 | ↓ 8 for rest of day. | Not noted |
| Fatal pneumonia | 17 | Strychnin, gr. 1/5 subcutaneously. | 0 | 0 | ↑ 10-30 for one hour | ↑ 15 for one-fourth hour |
| Infectious Colitis | 18 | Strychnin, gr. 1/10 subcutaneously. | ↑ 10 for one-fourth hour. | 0 | 0 | Not noted |
| Same case | 18 | Sterile $H_2O$ subcutaneously one hour later. | ↑ 10-12 for twenty minutes. | ↑ 5-10 for twenty minutes | ↑ 5 for one hour | Not noted |
| Delirium tremens | 19 | Strychnin, gr. 6/10 subcutaneously in six hours. | ↑ 15 after first injection, then falling to original level. | Could not be accurately read. | ↓ 5-8 for two hours | 0 |
| Fatal pneumonia | 20 | Strychnin, gr. 1/10 subcutaneously. | 0 | 0 | 0 | ? |
| Fatal tuberculous enteritis | 21 | Strychnin, gr. 1/10 subcutaneously. | ↑ 10 for one-half hour. | Could not be accurately read. | 0 | 0 |
| General peritonitis | 22 | Strychnin, gr. 1/40 subcutaneously at 9:18 and 10:50 a. m. | 0 | Could not be accurately read. | 0 | 0 |
| Surgical shock; recovery | 23 | Strychnin, gr. 1/10 subcutaneously. | 0 | Could not be accurately read. | 0 | 0 |
| Severe typhoid | 24 | Strychnin, gr. 1/10 subcutaneously. | 0 | 0 | 0 | 0 |

*↑ rise or increase; ↓ fall or decrease.

†The systolic pressure three days later at the same time of day was only 8 mm. less than the greatest height following strychnin, and 20 higher than it had been the previous morning.

TABLE 2.—Showing Effect of Caffein-Sodio-Salicylate in Single and Repeated Doses on Systolic and Diastolic Blood-Pressure, Pulse and Respiration

| Type | Case No. | Treatment | Systolic Pressure. Duration of Rise or Fall * | Diastolic Pressure. Duration of Rise or Fall * | Pulse. Duration of Increase or Decrease in Rate * | Respiration. Duration of Increase or Decrease in Rate * |
|---|---|---|---|---|---|---|
| Fatal lobar pneumonia | 15 | Caffein, gr. 1½ subcutaneously. | 0 | 0 | 0 | 0 |
| Fatal lobar pneumonia | 17 | Caffein, gr. 1½ subcutaneously each two hours for three days. | 0 | 0 | 0 | 0 |
| Fatal lobar pneumonia | 20 | Caffein, gr. 1½ subcutaneously each two hours for five doses. | ↓ 35 during these ten hours | ↓ 10 during these ten hours. | ↑ 10 during ten hours. | 0 |
| General peritonitis | 22 | Caffein, gr. 5 subcutaneously. | 0 | Could not be accurately read. | 0 | 0 |
| Fatal tuberculous enteritis | 21 | Caffein, gr. 5 subcutaneously. | 0 | Could not be accurately read. | 0 | 0 |
| Traumatic shock | 23 | Caffein, gr. 5 subcutaneously. | 0 | Could not be accurately read. | 0 | 0 |
| Same case | 23 | Scrubbing wound with tinct. iodin. | ↑ 20-30 permanently. | ? | 0 | 0 |
| Fatal septicopyemia | 25 | Caffein, gr. 5 subcutaneously. | 0 | 0 | ↓ 8 for less than fifteen minutes | 0 |
| | 25 | Caffein, gr. 5 subcutaneously at 10:50, followed by gr. 3 at 11:50, ten days later. | 0 | 0 | 0 | 0 |
| Same case | 25 | Caffein, gr. 17 in eight hours. | Not observed | Not observed | 0 | 0 |
| Surgical shock | 26 | Caffein, gr. 5, subcutaneously. | 0 | 0 | 0 | 0 |

*↑ rise or increase; ↓ fall or decrease.

tions, variations in the readings of less than 10 mm. could only be obtained in one case out of five. Our readings were made under conditions far from ideal. The patients were nearly all exposed to the noise and bustle and changing temperature of an open ward. They were all sick, some of them desperately so, some of them had painful diseases, others were receiving subcutaneous medication every hour. It is not necessary to name over every external condition which might have had an influence on the pressure curve in these individuals. Under such conditions, variations of 10, 15 or even 20 mm. are to be expected as the result of unavoidable external stimuli, and unless very consistent, cannot be held up as evidence in favor of the view that the drug is increasing the activity of the vasomotor center, or that any other effect peculiar to the drug has been produced. The same reasoning applies to the rate of the heart-beat.

It is important to point out that the measurements we have made do not permit us to state directly whether the drugs examined increase the efficiency of the circulation. We are justified in doing nothing more than make cautious inferences about the matter from the data obtained. It is true that a very rapid pulse is an ominous sign in the acute infections and that an agent which can reduce the rate of the heart-beat under such conditions must be considered a valuable therapeutic possession. On the other hand, the fact that the pulse is not slowed by any given drug, does not necessarily mean that the drug may not be improving the circulation in other ways. It is equally true that rises of blood-pressure do not necessarily signify better circulation, and that an increased flow of blood may occur in the absence of any significant change in general blood-pressure.[18] It has been shown by Hooker[19] that increasing the pulse-pressure in the renal artery is followed by increased secretory activity on the part of the kidney, but it must be remembered that measuring the pulse-pressure in the brachial artery does not necessarily tell us anything about the pulse-pressure in the kidney or heart and that local changes in pulse-pressure are, in all likelihood, occurring in the organs according to the state of their activity.

With these limitations in mind, we present our data in Tables 1 and 2, and in the accompanying clinical histories and charts, and offer the following summary of the observations we have made.

18. See for example, Price, F. E.: Brit. Med. Jour., 1912, p. 689, and Sollmann and Pilcher, *loc. cit.*, Footnote 16.

19. Hooker, D. R.: The Influence of Pulse-Pressure on Renal Function, Am. Jour. Physiol., 1910, xxviii, 24.

## SUMMARY

### STRYCHNIN

1. Eight times out of seventeen there was no change in systolic pressure. Once it fell 10 mm. for two hours. Nine times out of seventeen there was a rise in systolic pressure. This rise was small, usually brief, often of only a few minutes duration. It was inconstant, never to be relied on, often occurring after the first injection of a series, then falling to the original level in a short time and not recurring after subsequent injections. In no instance was the rise accompanied by improvement in the clinical condition. Only once was there a rise of more than 15 mm. In this single instance the pressure went up 26 mm. for a few minutes. In one patient the examination of the lungs caused a rise of from 20 to 30 mm. which lasted one hour; in another patient, subcutaneous injection of water caused a greater rise than $\frac{1}{10}$ gr. strychnin.

2. Nine times out of thirteen there was no effect on the diastolic pressure. Once it fell 10 mm. for forty minutes. In another case 15 mm. for one and one-half hours. Once it rose 30 mm. for fifteen minutes.

3. Eleven times out of sixteen the pulse showed no change. In no case was there a slowing which could be honestly attributed to the drug.

### CAFFEIN

1. Eight out of nine times there was no effect on the systolic pressure. Once it fell 35 mm. during the period of treatment. There was not a single rise in systolic pressure as a result of administering caffein.

2. Five out of six times there was no effect on diastolic pressure. Once it fell 10 mm. during the period of treatment.

3. Nine out of ten times there was no effect on the rate of the pulse. Once it rose 10 beats per minute during the period of administration.

4. In no instance was there a measurable change in respiration.

A few cases deserve special comment. The failure of a large dose of strychnin, gr. $\frac{1}{10}$, to raise the pressure of a patient with tuberculous enteritis whose systolic pressure had been 80 to 95 mm. for several weeks, was considered highly significant.

A patient with delirium tremens, a frail, small woman, received $\frac{1}{10}$ gr. of strychnin every hour for six hours, and after an interval of seven hours three more doses at the rate of $\frac{1}{10}$ grain per hour, without showing either toxic symptoms or any blood-pressure change.

Case 23 is one of typical surgical shock induced by fracture of the skull. His initial systolic pressure, which was 55 to 65 mm., was unaffected by either 1/10 gr. strychnin or 5 gr. of caffein-sodio-salicylate. Nevertheless the failure of these drugs to show a pressor effect cannot be explained by assuming that the function of the vasomotor center was in abeyance, for stimulation of the peripheral nerves was followed by a prompt and lasting rise in the pressure of 30 mm. The stimulus consisted of scrubbing the wound with tincture of iodin. A similar condition was met in Case 12, in which two 1/10 gr. doses of strychnin, given within an hour, had no effect on the falling blood-pressure, but physical examination was followed by a rise of 40 mm.

## *CONCLUSIONS*

1. There is no evidence that the vasomotor apparatus is injured in the acute infectious diseases.

2. The evidence at hand does not permit us to say whether the functional activity of the myocardium is seriously impaired in the acute infectious diseases.

3. Strychnin sulphate, in medicinal doses, does not increase the output from the heart, slow the pulse or materially raise the blood-pressure. There is no logical basis for its use as a cardiovascular stimulant.

4. Caffein-sodio-salicylate, in the doses employed, does not raise the blood-pressure or slow the pulse. The method does not permit us to say whether caffein increased blood flow in the cases studied.

## CASE HISTORIES

Case 9.—L. S., Pernicious anemia. Hgb. 30 per cent. Red cells 1,500,000. The patient complained chiefly of great weakness and anorexia. His temperature was slightly elevated and his pulse somewhat rapid. Examination of the heart showed the apex beat to be in the fifth space 3¼ inches to the left of the midsternal line. The right border of dulness was 1¼ inches to the right of the midsternal line in the fourth space. The upper border was at the third rib. The sounds were regular and of good quality. At the apex there was a short systolic and a faint diastolic murmur. At the base similar murmurs were heard. The pulmonic second sound equalled the aortic second sound in loudness and neither was accentuated. The patient was started on strychnin and in the course of four days received 1 4/10 grains subcutaneously. There was no change in the cardiac or respiratory rate or in the blood-pressure. There was certainly no improvement in the subjective symptoms, and the patient lost 3 pounds during this period. The physical examination of the heart was as before the strychnin period.

Case 10.—D. C., aged 23, typhoid fever. Moderately sick. Not in need of stimulation. Uneventful convalescence. On the third hospital day, 1/20 gr. of strychnin was followed by a rise in systolic pressure from 108 mm. to 118 mm. Two days later 1/10 gr. of strychnin was followed by a rise in pressure from 108 mm. to 115 mm.; 1/10 gr. of strychnin had no more effect than 1/20 gr. In both instances the systolic pressure was raised 10 mm. for less than one

hour. In the first instance the diastolic pressure did not change, consequently the pulse pressure was increased 10 mm. In the second instance the diastolic pressure fell 10 mm. with a resulting increase of 20 in the pulse-pressure. The rate of the pulse was not materially altered in either case. The strychnin caused no subjective sensations.

Case 11 (Chart 1).—A. M. S., aged 16 years. Typhoid fever. Mild attack. Not in need of stimulation. The rise in both systolic and diastolic pressures following the strychnin is striking, but it will be noted that the systolic pressure remained 20 mm. higher throughout the day than it had been just before the strychnin, and that on the next day it was about 110, which was 10 mm. higher than the initial pressure. Such a prolonged effect could scarcely be attributed to the strychnin. On the second day after the strychnin, the early morning systolic pressure was again 100 mm. but rose during the day to 110 mm.; the next day the early morning pressure was 120 mm. In brief, the systolic

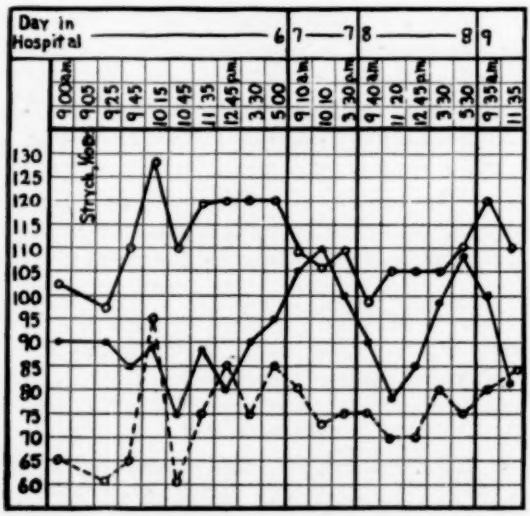


Chart 1 (Case 11).—In this and the following charts the temperature, pulse and respiration are represented in the usual way. The systolic blood-pressure is represented by circles connected by solid lines. The diastolic blood-pressure is represented by circles connected by broken lines. The dosage and the time of administration of the drug are printed on the chart (*Strych.* = strychnin sulphate).

blood-pressure of this boy, followed for three daws, showed a variation of twenty, due to influences of which we are ignorant. To attribute the rise of thirty on the first day entirely to the effect of strychnin would scarcely seem justified. It will also be noted that there was no increase in the pulse pressure following the giving of strychnin. The drug caused no subjective sensations.

Case 12 (Chart 2).—A. J. G., aged 39. Lobar pneumonia. Not in need of stimulation. On the morning of the third hospital day he was given strychnin, gr. 2/10, subcutaneously. Up to this time the systolic pressure had been running between 135 and 145 mm. and the diastolic pressure about 100 mm. After

the dose of strychnin the systolic pressure fell to 122 and the diastolic to 90 mm. The next morning at 8 a. m. the systolic pressure was still 120, the diastolic 85 mm. At 8:30 a. m. the patient was examined, and at 9 a. m. the systolic pressure was 150 and the diastolic 110 mm. One hour later the systolic pressure was still 140 mm. It is evident that large doses of strychnin had no effect on a falling blood-pressure, and it is equally apparent that mild exertion did have a well marked pressor effect.

Case 13.—P. T., aged 33. Lobar pneumonia. Not in need of stimulation. The systolic pressure had fallen progressively for the first three days in the

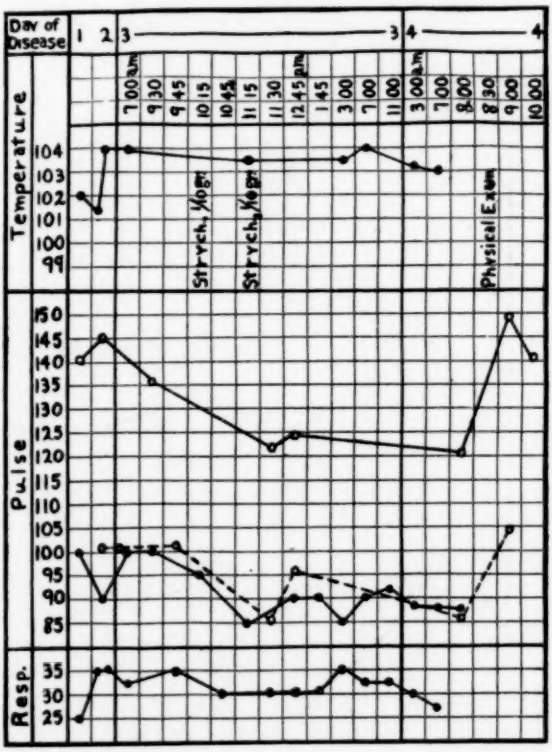


Chart 2 (Case 12).—Effect of strychnin sulphate (*Strych.*), as set forth in the case report.

hospital from 135 mm. to 120 mm. On the fourth day the patient was given strychnin sulphate, gr. 1/10, subcutaneously, at 12 noon and again at 1:30 p. m. No apparent change followed.

Case 14.—F. MacD., aged 32, lobar pneumonia. At 4 p. m. and again at 5 p. m. on the fifth day of the disease, and the third in the hospital, the patient was given strychnin sulphate, gr. 1/10, subcutaneously. He was doing well at the time and not in need of stimulation. The systolic blood-pressure was, however, 105 mm., and so the attempt was made to raise it by means of large

doses of strychnin. The latter had no effect on either the systolic or the diastolic pressure, or on the rate or character of the pulse.

CASE 15 (Chart 3).—L. C., aged 56. Lobar pneumonia, fatal on the fourth day in the hospital. The patient was extremely sick at entrance and failed steadily in spite of various therapeutic efforts. On the first day she was started on digipuratum, gr. 1½, three times a day, and camphor, gr. 3, subcutaneously,

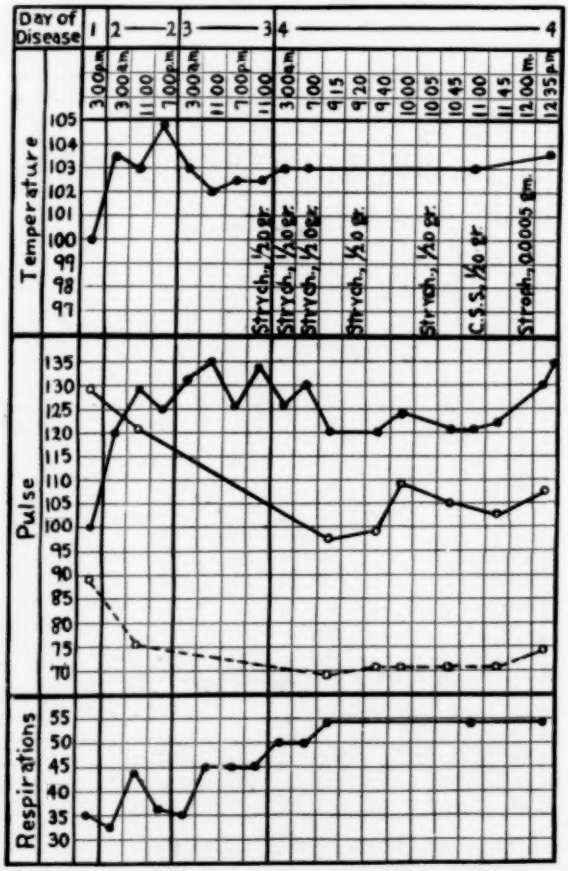


Chart 3 (Case 15.)—Effect of digitalis, caffein-sodio-salicylate (*C. S. S.*) and strychnin sulphate (*Strych.*) in a case of pneumonia.

each two hours, both of which were continued to the end. On the second day caffein, gr. 1½ each two hours was added. During the last twenty-four hours strychnin was given as shown on the chart. It will be seen that neither it, nor the camphor or caffein had any effect on the course of the disease.

CASE 16.—G. C. MacD., aged 25. Lobar pneumonia. Moderately sick. Not in need of stimulation. Systolic blood-pressure 110 mm., diastolic pressure

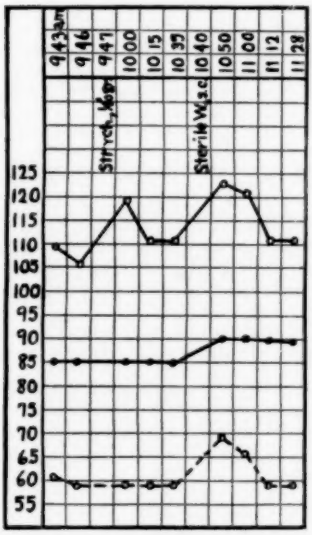


Chart 4 (Case 18).—Effect of strychnin sulphate (*Strych.*) in a case of chronic infectious colitis. *Sterile W. s. c.* = sterile water s. c.

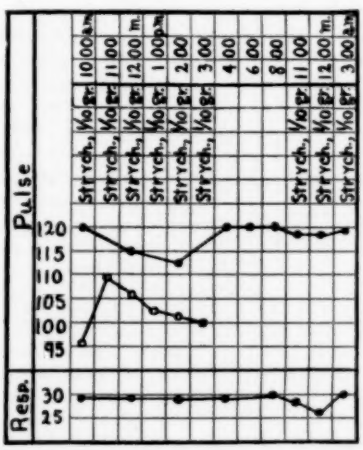


Chart 5 (Case 19).—Effect of strychnin sulphate (*Strych.*) in a case of fracture in an alcoholic.

80 mm. Strychnin sulphate gr. 1/20, subcutaneously, during the febrile period was followed by practically no change in the blood-pressure, but a slowing of the pulse, of twelve beats per minute. Strychnin sulphate, gr. 1/10, subcutaneously after the crisis, but while the pressure was still subnormal had practically no effect on the pressure or on the rate of the pulse. The strychnin caused no subjective symptoms.

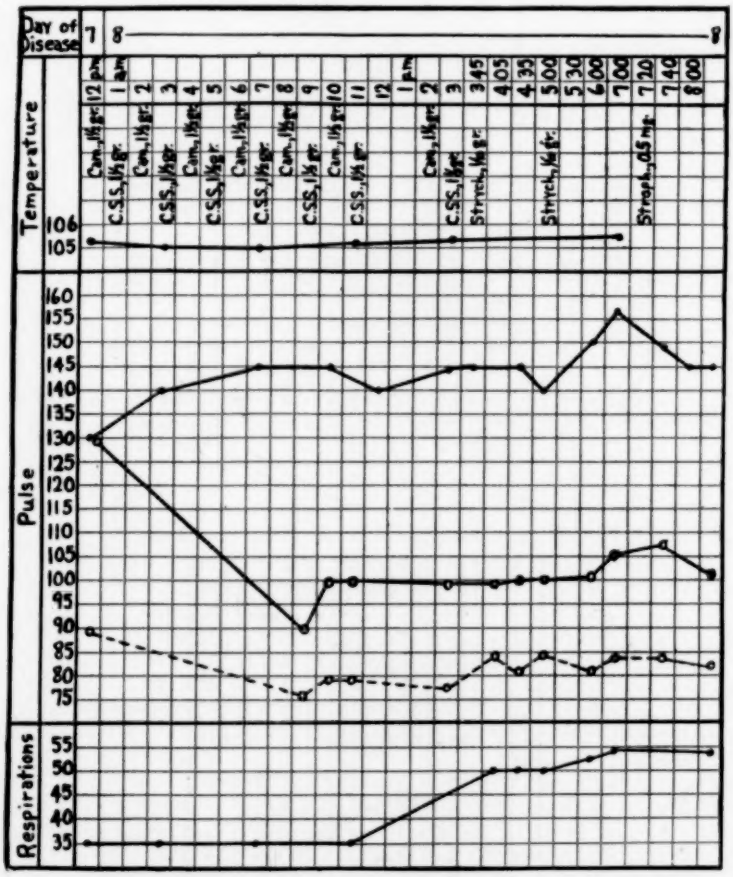


Chart 6 (Case 20).—Effect of camphor (*Cam.*) caffein-sodio-salicylate (*C. S. S.*) and strychnin sulphate (*Strych.*) in a case of lobar pneumonia. *Stroph.* = strophanthin.

CASE 17.—J. B., aged 47, lobar pneumonia. The patient died on the third day in the hospital. Tincture of digitalis, minims 15, three times a day; camphor, gr. 1½ each two hours; caffein-sodio-salicylate, gr. 1½ each two hours, alternating with the camphor, were given throughout the illness. On the third

day in the hospital when the systolic pressure was 110 mm. and the diastolic 75 mm. he was given 1/5 gr. of strychnin. No perceptible results followed.

Case 18 (Chart 4).—C. A., aged about 25. Chronic infectious colitis. Marked loss of weight. Marked weakness. Not in need of stimulation. Following 1/10 gr. of strychnin the systolic pressure rose 12 mm. for fifteen minutes. The diastolic pressure did not change. A hypodermic injection of water was followed by a greater and somewhat more prolonged rise in systolic pressure, and a rise of 10 mm. in diastolic pressure.

Case 19 (Chart 5).—N. R. A., aged 51. Fracture of the femur. Alcoholic history. Active delirium tremens. The chart shows what large amounts of strychnin may be given without producing any material or lasting change on systolic blood-pressure, pulse-rate, or respiratory-rate. The patient had 6/10 gr. of strychnin in six hours and a total of 9/10 gr. in seventeen hours without showing any toxic symptoms or even a slowing of the pulse.

Case 20 (Chart 6).—F. O., aged 41. Fatal lobar pneumonia. Several hours after entrance to the ward the systolic pressure was 130, the diastolic pressure 90 mm., and the pulse-pressure 40. The next morning the systolic pressure had

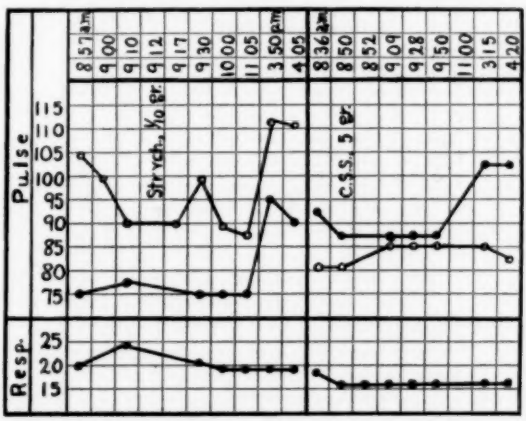


Chart 7 (Case 21).—Effect of strychnin sulphate (*Strych.*) and caffein-sodio-salicylate (*C. S. S.*) in a case of tuberculous enteritis and peritonitis.

fallen to 90-100, the diastolic to 75-80, and the pulse-pressure to 15-20 mm. Hg. During the night she had received camphor, gr. 1½ subcutaneously each two hours for five doses, and caffein, gr. 1½ each two hours for five doses. Between 10 a. m. and 2 p. m., and again between 2 p. m. and 6 p. m. she had had no camphor. There was, however, no fall in blood-pressure or rise in pulse. Between 11 a. m. and 5 p. m. she had had but one dose of caffein. There was, however, no fall in blood-pressure or rise in pulse. This would seem to indicate that neither the blood-pressure nor the pulse had maintained the level recorded earlier in the day because of the administration of the drugs. At 3:45 p. m. she was given strychnin, gr. 1/10 subcutaneously without effect on the pulse or the blood-pressure. Following strophanthin at 7:20 p. m. there was a slight slowing of the pulse and a slight rise in blood-pressure. The patient died at 3:30 the next morning.

Case 21 (Chart 7).—A. X., aged about 32. Tuberculous enteritis and peritonitis. Diarrhea of four months' duration. Great loss of weight. Marked weakness and emaciation. Irregular afternoon rises of temperature. The sys-

tolic blood-pressure was 85-95 mm. in the morning with the patient in bed for several days preceding the injection of strychnin. The diastolic pressure could not be determined by the auscultatory method. The curve shows well the initial rise which often follows the application of the apparatus, and also the normal afternoon rise in blood-pressure. The injection of 1/10 gr. of strychnin was without any definite effect. Several days later, when the systolic blood-pressure was 80-85 mm., the injection of 5 grains of caffein-sodio-salicylate was without effect on the systolic pressure, the rate of the heart, the respiration or the subjective sensations.

CASE 22.—E. B., aged 10 years. Gangrenous appendicitis. Operation. Peritonitis. Bronchopneumonia. On the second day after operation the boy was in poor condition and the attending surgeon thought he had about an even chance for recovery. The systolic blood-pressure showed a peculiar phenomenon—the first beats at the wrist were felt at 115 mm. These beats occurred during expiration. During inspiration the pulse disappeared regularly and could not

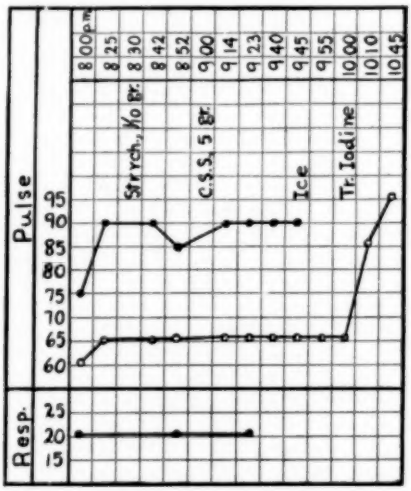


Chart 8 (Case 23).—Effect of strychnin sulphate (*Strych.*) and caffein-sodio-salicylate (*C. S. S.*) in a case of shock following an accident.

be felt until the mercury column had fallen to 90, when all the radial beats could be felt. Two doses of strychnin sulphate, gr. 1/40 subcutaneously, had no effect on this phenomenon, or on the rate of the pulse, which was 140, or on the rate of respiration, which was 35. Later in the day 5 gr. caffein were given without effect. On the fifth and sixth days his general condition was about the same, but his blood-pressure was falling. On the seventh day when the maximum systolic pressure was 95 mm. the pulse-rate 120-130 and the respiration 30, he had caffein, gr. 3 subcutaneously, every two hours for five doses without any apparent effect on the blood-pressure. The pulse, however, rose to 140-150 and the respirations to 40 during the caffein period. He recovered after several weeks of illness.

CASE 23 (Chart 8).—W. MacC., aged 27. March 26, 6 p. m., while crossing a railroad track he was struck by a train. He was brought to the hospital in shock at 7:10 p. m., and immediately placed on the "shock table." The acci-

dent had resulted in a fracture of the skull in the left frontal area. The patient was conscious but lethargic and in no pain. He was able to give his name. His skin was cold and colorless and his lips were blue. The systolic blood-pressure was 55-65 mm. Pulse was small but regular and slow. His heart sounds were not remarkable. His pupillary reflexes and knee-jerks were present. Neither strychnin sulphate, gr. 1/10 subcutaneously, nor caffein-sodio-salicylate, gr. 5 subcutaneously, had any effect on the blood-pressure, pulse or general condition. At 9:45 p. m., about two and one-half hours after being in the hospital, a piece of ice was placed on his abdomen for thirty seconds to see whether marked external stimuli (cold) would have any effect on the systolic pressure. None was observed. At 10 p. m. his wound was scrubbed with tincture of iodin.

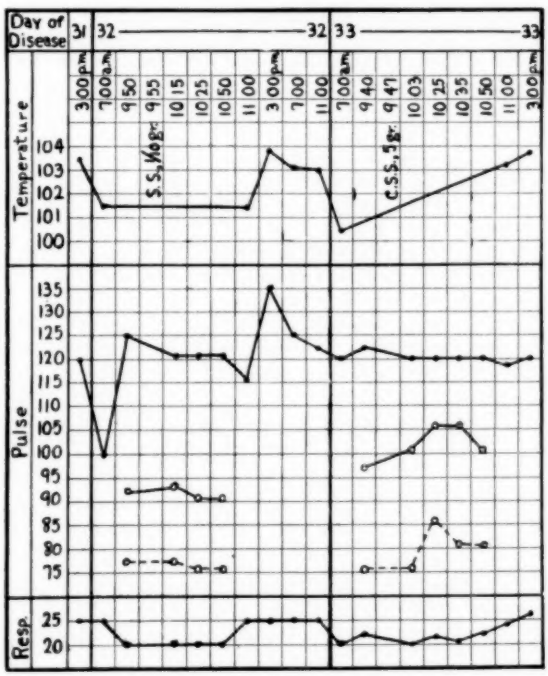


Chart 9 (Case 24).—Effect of strychnin sulphate (*S. S.*) and caffein-sodio-salicylate (*C. S. S.*) in a toxic case.

This caused a great deal of pain and resulted in an elevation of the systolic pressure, at first to 85 and then to 95 mm. This rise in pressure was taken to mean that the vasomotor center was not paralyzed, but was still capable of responding to stimuli, if sufficiently great, and that consequently the failure of the drug to cause a response could not be explained by the assumption that the center was paralyzed. The next morning the patient had entirely recovered from the shock. He was rational and alert and expressed a great desire for food. His systolic pressure was 95 and his diastolic 65 mm.

Case 24 (Chart 9).—P. M., aged 49. September 15, entered the hospital after one week's illness. September 27, very toxic. September 30, still more

toxic; low muttering delirium. October 5, the pulse was recorded at 170 for several hours. No hemorrhage at any time. October 16 and 17, strychnin and caffein were given. The strychnin was followed by practically no change in the cardiac or respiratory rates or in the blood-pressures. Caffein was without effect on the cardiac or respiratory rates and the very moderate changes in the systolic and diastolic pressures resulted in a lessening of pulse-pressure. The pulse showed a well marked respiratory rhythm and a slight irregularity in volume. Neither of these qualities was affected by the drugs. The patient had been very sick for some days preceding October 16, but was not losing ground and was not in direct need of stimulation when the above mentioned drugs were administered.

CASE 25.—J. G., aged 24. August 20, fever, rapid pulse, leukocytosis, multiple pyemic abscesses. Operation. Did not pick up following operation. September 6, right shoulder joint incised. September 10, growing worse. No hope for recovery. From September 5 to 8, caffein-sodio-salicylate, gr. 2, each two hours subcutaneously, and camphor, gr. 3, each two hours subcutaneously, had been given. September 8 the camphor was omitted, because it was thought that it was doing no good, and the caffein was reduced to gr. 1. September 11, blood-pressure readings were started. September 12, administration of 5 grains of caffein was without effect on the blood-pressure, the rate of pulse, or the rate of the respiration.

September 21. Since the last note the patient has been failing steadily. He looks as if he were about to die. Caffein, gr. 5, followed by gr. 3, was without effect on the blood-pressure. The pulse, however, fell 25 beats. This fall can scarcely be attributed to the drug, because the pulse had been at the lower level earlier in the morning. Shortly before the 10:45 a. m. reading, a painful dressing had been done. The pulse-rate increased in response to this and in the natural course of events it fell again to its previous level. Caffein was purposely given at this time to emphasize the care that must be exercised when judging of the effect of a drug under these complex conditions. The respiration became distinctly slower, following the administration of the drug, and there is no evidence to show that this was not a caffein effect. September 22, the patient received 17 grains of caffein in eight hours. It was without apparent effect on the respiratory or cardiac rates. September 23, died.

CASE 26.—A. P., aged 40. Traumatic compound fracture of right tibia and fibula. Fracture of right femur. He was brought to the hospital at 9:45 a. m., two hours after the accident, in surgical shock. He was given morphin sulphate, gr. ¼ subcutaneously, on entrance. The systolic blood-pressure was 85 mm., diastolic 50 mm., pulse 62, and the respiration 18. At 10:35 a. m., 5 gr. of caffein were injected. No measureable effect followed.

# IMMUNITY TESTS IN COCCIDIOIDAL GRANULOMA*

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Human infection by the organism now known as *Coccidioides immitis* was first reported in 1892 by Posadas and Wernicke in Buenos Aires. These observers regarded the parasites as protozoa, but later (1896) Rixford and Gilchrist gave the organism the binominal designation of *Coccidioides immitis*, and Ophüls succeeded in obtaining pure cultures in 1900. Other cases have been recognized from time to time, and recently MacNeal and Taylor[1] have collected twenty-four cases from the literature. In one of these reported by Brown, however (Case 18), the diagnosis was not confirmed culturally or by microscopic examination. All the reported cases except the first have occurred in the United States, and in a large proportion of them the infection can be traced to the San Joaquin Valley in California. The disease affects chiefly adult males, only one woman and one child being included in the cases observed. Only two patients are known to have recovered.

The infection is characterized by the formation of successive nodular or suppurative foci which may affect any part of the body, but more often are localized in the bones and joints, lymph-nodes, lungs and skin. The clinical symptoms depend on the anatomical distribution of the lesions. Histologically, the process resembles tuberculosis and belongs to the group of the infectious granulomata, the disease usually being termed, therefore, coccidioidal granuloma. In sections and in pus from the lesions the organism occurs as refractile, doubly-contoured, spherical bodies varying from 3 to 50 microns in diameter. These differ from the parasites found in blastomycosis in that budding forms are never seen in the animal body. In the tissues *Coccidioides immitis* multiplies by endosporulation and mycelium is never found, but when transplanted on artificial culture mediums the spherical bodies form mycelium almost immediately. Growth appears usually in less than a week and is more rapid at incubator temperature. Preparations from cultures show septate branching hyphae on which after a period of time chlamydospores develop. Dogs, rabbits and guinea-pigs can easily be infected with pus from one of the lesions or with emulsions from cultures. For more details concerning the clinical, pathological

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* From the Pathological Laboratory of the University of California Hospital.

1. MacNeal and Taylor: Jour. Med. Research, 1914, xxx, 261.

and microbiological features of the infection reference may be made to the papers of Ophüls,[2] Wolbach,[3] Hektoen,[4] Brown[5] and MacNeal and Taylor.[1]

The purpose of this paper is to add two other cases to the list of those already observed, and to report the results of a series of experiments designed to demonstrate specific immune substances in the blood of one of these patients.

## CASE REPORT

*History.*—A Portuguese man (M. M.), aged 25, was admitted to the surgical wards of the University of California Hospital, Dec. 1, 1913, with multiple abscesses. He was born in the Azores Islands, but had been in this country for many years. During the three months preceding his present illness he had been employed as a milker on a ranch near Newman, Cal. (San Joaquin Valley). His family history and past history were both negative. He remembered no previous illnesses and had not been associated, so far as he knew, with anyone affected with ulcerations on the skin or with abscesses.

The history of his illness follows: About nine months previous to admission he noticed a painful lump on his right wrist. This was opened by a physician and exuded pus for a considerable length of time. Later his right knee began to swell and after three months suppurated. It was aspirated by another physician and placed in a plaster cast, but as the rapid suppuration continued he was removed to a hospital and the joint was resected. The wound did not heal after this operation but continued to discharge pus. During this time other suppurative foci appeared over the right shoulder, left arm, left ankle below the external malleolus, and two foci beneath the scalp.

*Physical Examination.*—On admission (Dec. 1, 1913) the right knee showed numerous sinuses and the resected joint was mobile and painful; the right foot showed a sinus at the first distal metatarsal joint, running up the inner side of the foot to an ulcer over the Achilles tendon; the right wrist was riddled with sinuses discharging pus, the carpus seemed disintegrated and the lower end of the radius was swollen; left foot showed a discharging sinus below the outer malleolus, the left ulna was thickened and just below its middle showed a painful soft fluctuating mass about 6 cm. in diameter; two sinuses in the scalp ran down to the skull, one over the frontal bone just to the right of the median line and the other about 7 cm. posterior to the first; the left knee joint was distended and painful. The ulcerated areas at the sinus openings showed flat irregular granulations at the edges and were bathed in a purulent secretion.

The patient appeared considerably emaciated and was quite weak. Physical examination was otherwise negative except for a moderate enlargement of the cervical, axillary and inguinal lymph-nodes and a slightly impaired resonance at the left pulmonary apex.

*Course.*—The left knee gradually became more swollen and was aspirated several times. December 24 it was opened and 1,000 c.c. of pus removed. During the next two months the patient became weaker and more emaciated, all the lesions continued to discharge abundant purulent material, and new suppurative foci appeared on the left hip, right axilla and left shoulder.

March 8 the patient left the hospital against advice. He died a few days later and no necropsy was permitted.

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2. Ophüls: Jour. Am. Med. Assn., 1905, xlv, 1291.
3. Wolbach: Jour. Med. Research, 1904, xiii, 53.
4. Hektoen: Jour. Am. Med. Assn., 1907, xlix, 1071.
5. Brown: Jour. Am. Med. Assn., 1907, xlviii, 743.

During his stay in the hospital his temperature ranged between 37 and 38 C., and the leukocytes varied from 12,000 to 20,000; hemoglobin was 46 to 60 per cent. (Dare); urine was negative for sugar and albumin. Blood-cultures taken on three occasions were negative. Wassermann was negative. The spherical doubly-contoured organisms were demonstrable in fair numbers in every specimen of pus examined. One specimen of sputum was reported to contain coccidioides, but this could not be verified as no more sputum could be obtained.

*Medication.*—Two doses of neosalvarsan, 0.6 gm., were given intravenously and also large doses of potassium iodid, without any apparent effect.

*The Parasite.*—Numerous examinations of pus from the various lesions showed pus cells and rather numerous refractile doubly-contoured spheres which varied considerably in size; some were no larger than a red blood corpuscle, while the largest one measured was 40 microns in diameter. All gradations between these sizes were seen. The spheres were for the most part composed of a coarsely granular protoplasm in which little structure could be distinguished. Some cells, however, appeared filled with small spheres each of which had a sharply defined individual capsule, and occasionally the smaller bodies could be seen escaping from one of these sporangium-like forms. No budding forms were ever observed.

Cultures at incubator temperature showed a definite tenacious growth at the end of twenty-four hours, which after a few days spread over the whole surface. Old cultures, especially those kept at room temperature, developed abundant aerial hyphae and appeared fluffy. In block preparations on glucose agar, the spore-like bodies from the pus at the end of twenty-four hours showed thread-like outgrowths of mycelium which later became branched and segmented. In older cultures the extremities of the segments became bulbous or clubbed and chlamydospores developed on some of the hyphae.

Male guinea-pigs were inoculated both with a few drops of pus from one of the lesions and with a small amount of an emulsion of a two-months-old agar culture. After a few weeks there occurred in both cases the typical marked enlargement of the testes and the animals were killed. At autopsy suppurative foci were found in the testes, lungs and liver, from which the spherical doubly-contoured bodies were readily demonstrable and pure cultures were recovered.

The patient was an Italian man aged 29, and the history obtained was very meager. Nine months previous to his admission, while at Bakersfield (San Joaquin Valley), he noticed a swelling of the cervical glands. He had been treated for some time in a tuberculosis sanatorium.

Physical examination showed an emaciated man who breathed with difficulty on account of large nodular masses in the neck. On the forehead above the left eye was a large sinus about 4 cm. in depth from which thick yellowish pus was discharging.

At the base of this sinus was necrotic bone. On the left shoulder were two discharging sinuses which extended beneath the clavicle and a similar open sinus was present in each supraclavicular fossa. In the neck extending downward from the angles of the jaw and in the posterior cervical triangles could be felt large, somewhat softened nodular masses. Both lips showed several healed scars. Aside from the special lesions noted, physical examination was negative. On the fourth day after admission the patient died from asphyxiation. Examination of pus from the discharging lesions showed numerous refractile spherical, doubly-contoured bodies, varying somewhat in size, but as a rule about three times as large as a pus cell. Many of these spheres were apparently filled with numerous small spore-like bodies. No budding forms could be found. Cultures gave a mold-like growth.

An additional case of coccidioidal granuloma was observed in July, 1914, at the Santa Clara County Hospital, San Jose, Cal., by Dr. Elizabeth G. Lewis, to whom I am indebted for the following clinical and post mortem notes:

At necropsy the numerous walnut-sized masses in the neck noted in the clinical examination were abscesses filled with yellowish pus, and there was also a large postpharyngeal abscess containing similar material. The apex of each lung was firmly bound by adhesions and formed part of the wall of an abscess which was discharging through a sinus above the clavicle. Section of the lung itself, however, showed no gross lesion and nothing noteworthy was noted in the examination of the rest of the viscera.

Tissue saved for microscopic study was unfortunately thrown away by mistake. The only material available for examination was an agar culture from the pus. This corresponded in all respects to the cultures of *Coccidioides immitis* from the case above described.

### ANTIBODIES

This patient was under observation for some time and attempts were made to ascertain what demonstrable antibodies were formed in response to the infection. The antigens used for this purpose were made from the strain of *Coccidioides immmitis* obtained from the patient and consisted of the two forms of the organism — (1) the mycelial growth from artificial culture mediums, and (2) the spore-like bodies from one of the purulent lesions. The mode of preparation of these antigens was as follows:

*1. Antigen from mycelial growth.*—A considerable number of agar cultures of *Coccidioides immitis* about four weeks old were used. The cotton plugs were treated with a few drops of formaldehyd solution and sealed. After twenty-four hours the tenacious growth was removed with a platinum spatula and dried over sulphuric acid. It was then powdered by grinding for several hours in an agate mortar.

*2. Antigen from the organism in the body.*—One thousand cubic centimeters of rather thick pus from an abscess (knee) was treated with 162 c.c. of antiformin and allowed to stand with occasional shaking for five hours. It was then centrifuged and the sediment washed five times with 0.85 per cent. NaCl solution, after which the reaction was neutral to litmus. Microscopic examination showed that the sediment was composed almost entirely of the spherical doubly-contoured bodies. Cultures remained sterile. This sediment was dried over sulphuric acid and the residue ground for several hours in an agate mortar. A second specimen of 500 c.c. of pus was similarly treated with antiformin, centrifuged and washed. A rather thick emulsion of the spherical bodies was made in 0.85 per cent. NaCl solution. This latter was used in the agglutination tests and was also used as antigen in some of the complement fixation tests.

*Controls.*—A similar powder was made in exactly the same manner from cultures of a strain of blastomyces from one of the Chicago cases. This organism showed many budding forms in culture in addition to mycelial growth.

Normal serums used in all the controls were from ambulatory patients not suffering from any infectious disease.

### COMPLEMENT FIXATION TESTS

The antigens used for these tests consisted of 0.5 per cent. emulsions of the powdered growth of coccidioides and blastomyces in 0.85 per cent. NaCl solution and in addition an emulsion of the spore-like forms of coccidioides obtained from the pus. A rabbit-antisheep

hemolytic system was used in all tests and the complement and hemolysin were always titrated before use. In the tests the inactivated patient's serum with antigen and 1½ units of complement were incubated for one hour and then two units of hemolysin (1:2,000) and 1 c.c. of a 5 per cent. suspension of sheep corpuscles added. The tubes were reincubated for two hours and placed in the icebox. The final readings were made the following day. Two normal serums were used as controls. On titration it was found that the emulsions of the dried mycelium of coccidioides inhibited hemolysis in amounts larger than 0.07 c.c., and that of blastomyces in amounts larger than 0.05 c.c., while the coccidioides "spores" did not inhibit in dilutions of 0.3 c.c. Varying amounts of serum up to 0.5 c.c. were tested and the results were entirely negative. Another similar series was carried out, using inactivated serum collected the same day the tests were done. This also gave negative results.

### CUTANEOUS REACTIONS

Since certain antigens like tuberculin, typhoidin and others cause a specific skin reaction, it was thought of interest to try similar reactions with coccidioides in the present case. Boughton and Stober[7] have reported an apparently specific skin test in a case of blastomycosis, although it is stated that the patient was recovering from the infection at the time the test was done. Davis,[8] however, was unable to obtain a positive skin reaction in guinea-pigs experimentally infected with a similar organism. In applying the tests both the intradermal method and scarification with a von Pirquet chisel were used. The following antigens were employed in each test: (1) A rather thick suspension of the powdered mycelial growth of coccidioides suspended in 0.85 per cent. NaCl solution; (2) a 5 per cent. glycerin emulsion of the same dried powder concentrated to one-tenth its original volume; (3) a concentrated glycerin bouillon filtrate from a four-weeks-old *coccidioides* culture prepared in a manner analagous to that used for making "old tuberculin"; (4) a rather thick suspension in 0.85 per cent. NaCl solution of the powdered spherical bodies from the pus; (5) and (6) suspensions of dried blastomyces growth prepared in a similar manner to (1) and (2). Controls of 0.85 per cent. NaCl solution and 50 per cent. glycerin were also used. A small drop of each of these was placed on a small area of scarification on the skin of the arm made with a von Pirquet chisel and allowed to dry. If the material had not dried at the end of five minutes, the excess was gently removed with absorbent cotton. No indication of any reaction appeared around any of

7. Boughton and Stober: THE ARCHIVES INT. MED., 1914, xiii, 599.
8. Davis: Jour. Infect. Dis., 1911, viii, 190.

the areas in the five succeeding days. The tests were repeated one month later and on two control cases with the same result.

In the intradermal tests, a very small amount of each of the antigens was injected in the superficial layers of the skin of the arm. In all except the controls and (3) the concentrated broth filtrate, there was formed an indurated red area of reaction from 5 to 10 mm. in diameter which at the end of forty-eight hours became pustular. The pus was sterile and the lesions had practically disappeared at the end of a week. This same reaction occurred in six control patients on whom the same tests were done and undoubtedly resulted from irritant substances present in the organisms themselves. The same tests were repeated after the various antigens had been diluted until no reaction was caused in control patients and the diluted antigens caused no reaction in the case of coccidioidal granuloma.

### AGGLUTINATION TESTS

The suspensions of coccidioides used in these tests were obtained as above described by treating a specimen of pus with antiformin, and consisted of a considerable number of the spherical doubly-contoured bodies in salt solution. The hanging drop method was used and two series of preparations made; one was incubated at 37 C. and the other left at room temperature. The dilutions made of the serum were 1 : 5, 1 : 10, 1 : 20, 1 : 40 and 1 : 80, and in each experiment the mixtures were made of (1) immune serum plus suspension, (2) normal serum plus suspension, and (3) salt solution plus suspension. Both the incubated series and that left at room temperature were examined at intervals of one hour for the first six hours, and again at the end of twenty-four hours. The results were entirely negative.

### PRECIPITIN TESTS

For these tests 0.3 gm. of the powdered mycelial growth of coccidioides was thoroughly ground with 30 c.c. of 0.85 per cent. NaCl solution to which 0.5 per cent. phenol had been added. This suspension was allowed to stand for several days, being shaken at intervals, and then centrifuged. The faintly opalescent supernatant fluid was employed in the tests. Similar extracts were made from the powdered coccidioides "spores" and from dried blastomyces cultures.

The first series of tests was carried out by stratifying the antigen above the serum in such a manner as to give a sharp line of contact. The tubes were incubated and readings made at the end of one hour and two hours. The various antigens were used in their full strength and also diluted 1 : 2, 1 : 5, 1 : 10 and 1 : 20, with 0.85 per cent. NaCl solution. As controls a solution of 0.85 per cent. NaCl solution with

0.5 per cent. phenol and normal serums were used. In all, serums from twelve control patients were tested. Table 1 gives the results of these experiments.

Another series of precipitin tests was carried out in the usual way by mixing various amounts of serum with the same antigens used in the previous experiment. The technic used was as follows: Varying amounts of clear serum from the patient with coccidioidal granuloma were added to a constant amount of the antigen extracts in small test-tubes and the mixture diluted to a uniform volume by the addition of 0.85 per cent. NaCl solution. The tubes were then incubated for two

TABLE 1.—RESULTS OF PRECIPITIN TESTS

| Extract, 0.5 c.c. | Serum from Case of Coccidioides, 0.5 c.c. | Normal Serum * 0.5 c.c. |
|---|---|---|
| Coccidioides culture | Heavy ring | No precipitate |
| Coccidioides 1 : 2½ | Distinct ring | No precipitate |
| Coccidioides 1 : 5 | Faint ring | No precipitate |
| Coccidioides 1 : 10 | No precipitate | No precipitate |
| Coccidioides 1 : 20 | No precipitate | No precipitate |
| Coccidioides "spores" | Distinct ring | No precipitate |
| Coccidioides "spores" 1 : 2½ | Faint ring | No precipitate |
| Coccidioides "spores" 1 : 5 | No precipitate | No precipitate |
| Coccidioides "spores" 1 : 10 | No precipitate | No precipitate |
| Coccidioides "spores" 1 : 20 | No precipitate | No precipitate |
| Blastomyces | No precipitate | No precipitate |
| Blastomyces 1 : 2½ | No precipitate | No precipitate |
| Blastomyces 1 : 5 | No precipitate | No precipitate |
| Blastomyces 1 : 10 | No precipitate | No precipitate |
| Blastomyces 1 : 20 | No precipitate | No precipitate |
| NaCl solution 0.85 per cent. + phenol 0.5 per cent. | No precipitate | No precipitate |

* Eleven other normal serums were tested with the same result.

hours at 37 C. and placed in the ice-chest. The readings were made at the end of twenty-four hours. The usual controls were introduced as may be noted from Table 2.

## SUMMARY

In a case of coccidioidal granuloma studied no specific complement-fixing bodies or agglutinins could be found in the blood-serum using cultures of *Coccidioides immitis* and emulsions of the same organism from human lesions as antigens. No specific skin reaction could be demonstrated. Precipitins, however, could be demonstrated in the serum even when diluted 1 : 160, when an extract of dried cultures of the organism was used as precipitinogen. These precipitins were apparently specific, since they could not be demonstrated when normal

serum was tested with the same antigen or when the specific immune serum was tested with an antigen similarly prepared from the closely related organism, blastomyces.

The presence of specific precipitins in this infection must be verified by the examination of other cases. It is suggested that this reaction might be applied as a means of diagnosis in cases of deep-seated infec-

TABLE 2.—Additional Precipitin Tests

| | Extract Coccidioides Culture | Extract Coccidioides "Spores" | Extract Blastomyces Culture | NaCl Solution 0.85 Pct. |
|---|---|---|---|---|
| Immune serum 1: 10 | + | + | 0 | 0 |
| Immune serum 1: 20 | + | + | 0 | 0 |
| Immune serum 1: 40 | + | + | 0 | 0 |
| Immune serum 1: 80 | + | 0 | 0 | 0 |
| Immune serum 1: 160 | + | 0 | 0 | 0 |
| Immune serum 1: 320 | 0 | 0 | 0 | 0 |
| Normal serum 1: 10* | 0 | 0 | 0 | 0 |
| Normal serum 1: 20 | 0 | 0 | 0 | 0 |
| Normal serum 1: 40 | 0 | 0 | 0 | 0 |
| Normal serum 1: 80 | 0 | 0 | 0 | 0 |
| Normal serum 1: 160 | 0 | 0 | 0 | 0 |
| Normal serum 1: 320 | 0 | 0 | 0 | 0 |
| NaCl solution 0.85 per cent. and phenol 0.5 per cent | 0 | 0 | 0 | 0 |

+ = precipitate; 0 = no precipitate.

* A series of nine other normal serums was tested with negative results.

tion where there are no discharging lesions from which the spherical doubly-contoured bodies can be demonstrated. It might also serve as a means of differentiating coccidioidal granuloma from blastomycosis in obscure cases.

Experiments are now being carried out to determine whether specific immune substances are formed in infected animals.

36 Farnsworth Lane.

# THE VALUE OF THE ELECTROCARDIOGRAM IN THE DIAGNOSIS OF CARDIAC HYPERTROPHY *

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The steady increase in the number of electrocardiographs, their constant clinical and experimental application, together with the resulting advance in the knowledge of cardiac activity, all go to show the need and importance of this method of investigation. The exactness of the instrument in the diagnosis of any of the arhythmias, and the simplicity of technic make it indispensable in any large clinic, where it tends to supplant the polygraph. When any new technical method is devised, be it in bacteriology, physics or diagnosis, it is at once applied to different problems in the constant effort towards the establishing of new truths. In such a way the electrocardiogram has come to be applied in an ever-widening field of cardiac investigation; and, among other uses, it has come to be a method for the determination of hypertrophy of the right and left sides of the heart. There must, of necessity, be a constant balancing of methods and results in order that the usefulness of a method be ascertained and that further progress be securely established.

To this end it was thought worth while to review the electrocardiograms obtained in the past few years at the Johns Hopkins Hospital, in an effort to compare the clinical findings, supplemented as they are by radiographic, fluoroscopic, and post-mortem knowledge, with the information obtained from the electrocardiogram. For this purpose some two hundred cases were taken, and their discussion will be the purpose of this paper. They include cases of clinical right- or left-sided hypertrophy with normal electrocardiograms, cases with apparently normal hearts with electrocardiographic findings of hypertrophy, and cases of apparently normal hearts from both the clinical and laboratory points of view.

At this point it would seem *apropos* to say just a word as to the physiological significance of the electrocardiogram. The different views, now current, only confirm the conception that the interpretation of the various peaks is by no means definite. Thus, is the "R" wave of papillary origin? Does the "T" wave result from contraction of a ring of muscle at the root of the aorta? Or, indeed, following Eyster's lead, does the "R" wave represent excitation and the "T" wave signify

* Submitted for publication Oct. 28, 1914.

contraction? To avoid a technical paper and to limit it to the clinical point of view, such conceptions will not be discussed. It will suffice to sum up the general opinion—to regard the "P" as of auricular origin and the "QRST" complex representative of ventricular activity, where any one point on the curve represents the algebraic sum for the particular derivation of the electrical potentials throughout the heart at that instant of time. The relative potential of the different sides of the heart change with the lead employed, so that the type of curve obtained in the left-arm and left-leg derivation is quite different from that obtained from the left-leg and right-arm. Furthermore, it would seem possible that different cardiac conditions—as relative hypertrophy of one side, change of the position of the heart—would be reason for changes in the ratio of the electrical potentials expressed by the electrocardiogram. Again, a thousand and one extra-cardiac conditions must be considered—the electrical conductivity of different tissues, the character of the retromanubrial tissue, the position of the body, the condition of the surrounding organs which might dislocate the heart. The age of the patient must be remembered, as the heart of a child is relatively larger than that of an adult, and then, with advancing age, varying degrees of sclerosis with varying cardiac responses develop.

Following hypertrophy of the left side of the heart, it is only too evident that a relative right-sided hypertrophy will result of an amount determined by the extent and duration of the original lesion and the reserve force on the right side. It would seem, then, that not only is the diagnosis of a pure left- or right-sided hypertrophy very difficult, but, a point much more fundamental, it is well nigh impossible to set up a physiological standard, and the term "normal" as applied to the cardiovascular system is very indefinite and, at best, is relative and varies with the individual. The standardization of the electrocardiogram based on such variability is difficult, and its diagnostic value decreases proportionately.

For the purpose of study, however, let us assume the general opinion that the normal electrocardiogram is one with no progressive increase or decrease in the size of the "R" wave in the three derivations—that left ventricular hypertrophy would give an "R" wave which would be shorter in "$D_2$" than in "$D_1$," and still shorter or inverted in "$D_3$." While the reverse—a progressive increase in the height of the "R" wave from "$D_1$" through "$D_2$" to "$D_3$"—is that associated with right ventricular hypertrophy. The absolute ignorance of the significance of the "Q" and "S" waves precludes their discussion.

At first in collecting normal cases, individuals of different ages were taken. They were non-patients and were presumably normal. But among these were found left- and right-sided hypertrophy, as well

as normal appearing electrocardiograms. This at once necessitated careful physical examination and a knowledge of their cardiac history; did they have athletic hearts, was there arteriosclerosis, or beginning chronic nephritis? To obtain a thorough physical examination and to avoid the personal element, it was thought fairer to cull from the hospital records non-cardiac cases that had electrocardiograms, and whose physical examination, made by different investigators, and often confirmed by roentgenography, or occasionally by post mortem, had disclosed normal sized hearts. Thus, fifty cases were obtained of clinically normal hearts, in which there was no apparent lesion of the cardiovascular system, and in which there was no demonstrable etiological factor which might lead to cardiac enlargement. These cases were diagnosed as neurasthenia, chronic appendicitis, gastric anacidity, and the like, and, in all of them, careful examination showed no cardiac abnormalities. Needless to say, that routine examinations by several men are more trustworthy than that made by one, and it would seem that such cases would be much more adaptable for the establishing of a norm than fifty persons taken off the street, so to speak, in whom the history and routine study would be much less complete.

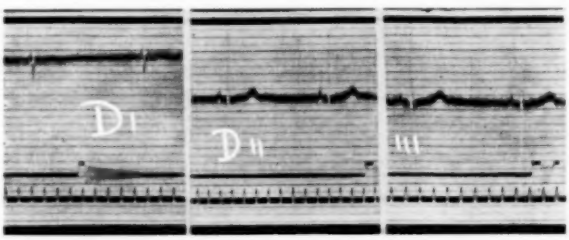


Fig. 1.—Right ventricular hypertrophy from the electrocardiogram with a clinically normal heart. Patient aged 15.

There were ten patients of ages varying from 10 to 20, who, as far as could be ascertained, were normal. Seven of these were boys from private schools who were taken in connection with heart-sound work, and whose physical examinations, checked by Dr. Thayer and Dr. Bond, were in no way remarkable. The other three patients of this age were hospital cases—dementia praecox and neurasthenia. Eight of these ten showed electrocardiographic findings of right ventricular hypertrophy. The others gave the normal sequence of derivations.

M. F., aged 15, diagnosed as having psychoneurosis, anorexia nervosa, dementia praecox, was a tall, thin girl weighing 55 pounds. She showed a normal venous-pulse. The apex was in the fourth interspace, 6 cm. from median sternal line, and the cardiac dulness extended 8.5 cm. to the left and not to the right of the median sternal line. The rest of the physical examination was

not remarkable, blood-pressure being 90, and the urinary findings normal. The Roentgen ray disclosed a heart in normal position and of normal size, which by actual measurement, corresponded closely to the clinical dimensions. The electrocardiogram as shown in Figure 1 is typically that of right ventricular hypertrophy.

Perhaps she did have a relative right ventricular hypertrophy, but it could hardly be regarded as pathological. That such a sequence of derivations is the normal at this age is also possible, but it is by no means constant, as two of the patients of this age did not show a progressive increase in the height of the "R" wave. The difficulty of

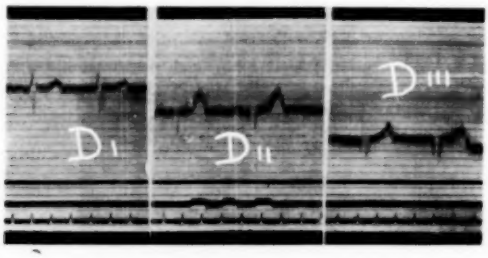


Fig. 2.—Right ventricular hypertrophy from the electrocardiogram with a clinically normal heart. Patient aged 14.

assuming a standard is at once evident, and without a standard the diagnosis of the pathological becomes, of necessity, indefinite. Six of the schoolboys showed the sequence of derivation associated with right ventricular hypertrophy, as seen in Figure 2, and one of the same age,

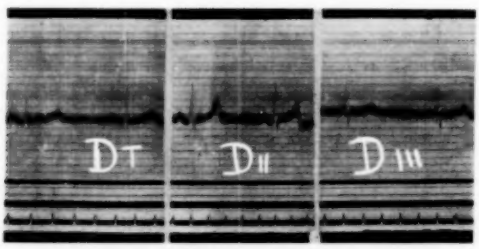


Fig. 3.—Normal sequence of derivation in a clinically normal heart. Patient aged 15.

again with normal cardiac findings, had, according to the electrocardiogram (Fig. 3) a normal relation of ventricular potentials in the three derivations.

In the same way there were fourteen whose ages varied from twenty to thirty—a group evidently useless in drawing statistical conclusions, but sufficient to show the lack of uniformity. Again the physical find-

ings were normal, and the diagnoses were psychoneurosis and neurasthenia. Eight showed normal electrocardiograms, three showed right-sided and three showed left-sided ventricular hypertrophy.

Mrs. L. H., aged 26, with a diagnosis of psychoneurosis, psychasthenia, and a questionable pulmonary tuberculosis, showed indefinite signs at the right base with a negative Calmette, and, according to Roentgen ray findings, some fibrosis of the lung. Her apex was in the fifth interspace, well within the midclavicular

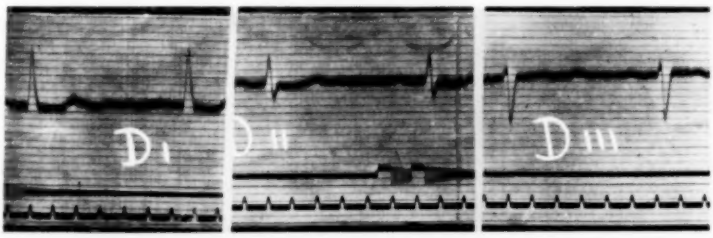


Fig. 4.—Electrocardiographic findings of left ventricular hypertrophy with a clinically normal heart. Patient aged 26.

line. Her cardiac dulness extended 3 cm. to the right and 9.5 cm. to the left of the median sternal line. Her blood-pressure was 125 and there were normal urinary findings. The Roentgen ray showed a normally placed heart of normal dimensions. The electrocardiogram of this patient (Fig. 4) showed marked left ventricular hypertrophy with "S" wave very marked in "$D_1$" and the "R" inverted markedly in "$D_3$." That is, a case of left ventricular hypertrophy according to the electrocardiogram, with, clinically, a normal heart.

It might be that this marked left ventricular hypertrophy is significant of early cardiac changes which would not be so prominent, as, with later developments, right ventricular changes appeared to cloak the present electrocardiographic signs. But, occurring as it does, with

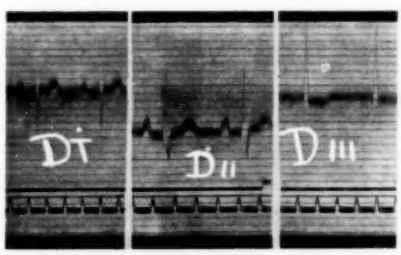


Fig. 5.—Electrocardiographic findings of right ventricular hypertrophy with a clinically normal heart. Patient aged 23.

no evident cardiac changes and where, from the possible presence of pulmonary tuberculosis, the expression of the first signs of cardiac hypertrophy would be expected on the right, this electrocardiographic

diagnosis seems questionable. Then at the same age, in a clinically normal heart there appear electrocardiographic changes of right ventricular hypertrophy.

W. J., a well built young man of 23, weighs 140 pounds. He was regarded as having a case of periodic paralysis. His cardiac dulness measured 4.5 cm. to the right and 9 cm. to the left of the median sternal line, with the apex in the fourth interspace, inside the nipple line. Blood-pressure and urinary findings were normal. Figure 5 shows his electrocardiogram.

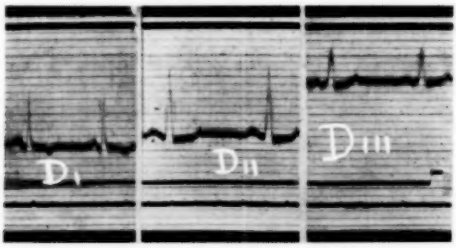


Fig. 6.—Normal electrocardiographic findings in a greatly hypertrophied heart which at post mortem was grossly much more marked on the left.

No further mention of the other eight cases need be made beyond the fact that they had clinically normal sized hearts with normal electrocardiograms.

Further discussion of the other decades emphasizes the absence of a standard, and suggests the conclusion that, while in youth the right ventricular hypertrophy type of electrocardiogram is a common one, as age increases the left ventricular type predominates. It is impossible to say that there may not actually be a true hypertrophy, but as there is no possible standard for the normal, how can the pathological be diagnosed, and of what use would be the electrocardiographic findings?

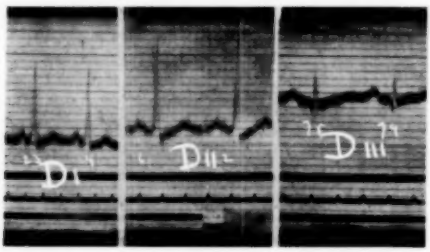


Fig. 7.—Normal electrocardiographic findings, where marked left ventricular changes would have been expected.

With this much information it will be interesting to review the electrocardiograms from a different angle—the comparison of all the

normal types of electrocardiograms with clinical findings, no matter what the cardiac condition. Sixty cases come under the broad conception of normal electrocardiograms — there was no progressive rise or fall in the size of the "R" waves. Of these, twenty-four would have been expected to be normal from the clinical findings, supplemented by the Roentgen ray. Twenty of the so-called normal electrocardiograms

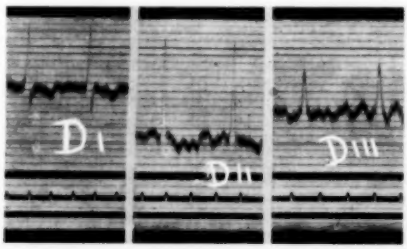


Fig. 8.—Normal sequence of derivations where right ventricular hypertrophy is suggested clinically. (Mitral stenosis).

were from cases that would, from clinical experience, have been regarded as definite instances of ventricular hypertrophy. Examples of this group seem advisable.

W. W., aged 49, a colored laborer of medium stature, weight 162 pounds, came into the hospital decompensated, and later died. The clinical diagnosis of myocardial insufficiency, aortic insufficiency, and arteriosclerosis was sub-

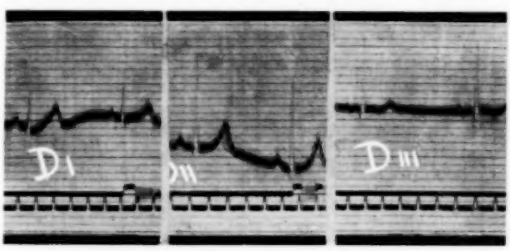


Fig. 9.—Normal electrocardiographic sequence in a case with lesions in both cardiac chambers.

stantiated at post mortem, and the heart, which had been greatly enlarged downward and outward with the apex in the sixth interspace, 12 cm. from the median sternal line, weighed 600 gm. There was marked dilatation and hypertrophy, especially of the left side, whose wall measured 2 cm. in thickness. There was right ventricular hypertrophy, but of an extent not to be compared with that of the left. The electrocardiogram, Figure 6, is that of a normal relation between the left and right ventricular potentials.

In the same way, A. B., a colored laborer, aged 60, was admitted and a diagnosis made of myocardial insufficiency, syphilis (Wassermann), arteriosclerosis, aortic insufficiency, dilated aortic arch. His apex was in the sixth

interspace and the precordial dulness extended 5 cm. to the right, and 13 to the left, which, for a man of 110 pounds, is a marked increase. His blood-pressure was 150 mm. and the urinary findings were normal. Surely it would be thought that this man had a marked left ventricular hypertrophy that was relatively much greater than any resulting right-sided enlargement. And yet, his electrocardiogram, Figure 7, was fairly normal in appearance.

Three of the cases which, clinically, should have been right-ventricular hypertrophy, showed normal electrocardiograms.

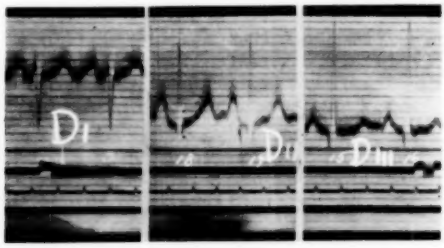

Fig. 10.—Electrocardiographic findings of right ventricular hypertrophy agreeing with the clinical findings.

T. H., a 45-year-old physician, showed typical signs of mitral stenosis and insufficiency, with symptoms of five years' duration following acute articular rheumatism. His apex was in the fourth interspace, 9 cm. from the median sternal line, with cardiac dulness extending to the right sternal border and 10.5 cm. to the left. The electrocardiogram (Fig. 8) shows, besides auricular fibrillation, a sequence of derivations associated with a normal relation between the two sides of the heart, in spite of the clinical findings which suggest right ventricular hypertrophy.

The remaining thirteen so-called normal electrocardiograms were obtained from patients with diseased hearts in whom, although it would have been expected that there would have been a predominating hypertrophy of one side or the other to give the electrocardiographic findings of hypertrophy, it is possible to conceive that there was a proportional hypertrophy of both sides to maintain a normal balance, and therefore a normal type of electrocardiogram.

F. S., aged 14, weighed 85 pounds, his cardiac symptoms were of eight years' duration, following an attack of acute articular rheumatism, and were diagnosed as resulting from mitral stenosis and insufficiency, with aortic insufficiency. Blood-pressure was 100, urine normal. The Roentgen ray showed a fairly normal sized heart. To percussion, the heart extended 5 cm. to the right, and 11.5 cm. to the left of the median sternal line, with the apex in the fifth interspace in the midclavicular line. His electrocardiogram (Fig. 9) was normal.

When one considers the tendency at this age to show normally a right ventricular sequence, and as this patient under normal conditions might have shown this sequence, it may be that in this normal sequence

we are dealing with an effort on the part of the electrocardiograph to express a pathological hypertrophy, a change from the normally found right-sided hypertrophy to the pathologically found normal sequence. Such hypotheses would seem to lead very far afield.

Among the collection of electrocardiograms, twenty-seven were found that suggested right ventricular hypertrophy. It would be advantageous to compare them with the clinical findings. Seven of these cases were clinically right-sided hypertrophy.

R. K., married, aged 28, weighing 94 pounds, with two years of cardiac symptoms, had a diagnosis made of pure mitral stenosis and exophthalmic goiter. To percussion, the heart measured 3.7 cm. and 11 cm. to the right and left of the median sternal line, with her apex in the left mammillary line. Blood-pressure was 85; urine, negative.

Probably this case was true right ventricular hypertrophy, and the electrocardiogram (Fig. 10) confirms this supposition. On the other hand, five cases of electrocardiographic right ventricular hypertrophy were found in patients with apparently normal hearts.

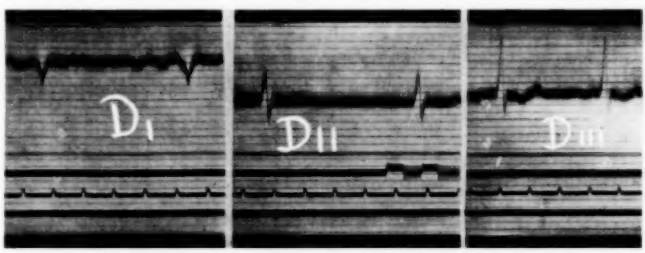


Fig. 11.—Electrocardiogram suggesting right ventricular hypertrophy in a clinical case of left ventricular hypertrophy.

It is striking to see four cases of electrocardiographic right-sided hypertrophy associated with clinical findings that point strongly to left-sided enlargement.

W. F., aged 51, weighing 124 pounds, was considered to have arteriosclerosis, myocardial insufficiency, aortic and mitral insufficiency. The Roentgen ray showed marked enlargement of the third curve on the left, signifying left ventricular enlargement. Blood-pressure was 100, urine showed chronic passive congestion changes. The patient came in badly decompensated and with auricular fibrillation. The heart, at the time of the electrocardiogram, measured 5.5 cm. to the right, and 14 to the left of the median sternal line. The apex was in the fifth interspace, three fingers' breadths outside of the nipple.

Quite possibly the patient did have a right ventricular hypertrophy, as the electrocardiogram (Fig. 11) showed, and as might be expected from the presence of decompensation. But if these electrocardiographic findings result from the preponderance of the right side, it is hard to believe that, with the Roentgen-ray and clinical findings, the

right side should have reacted more to the original lesion than did the left.

The eleven remaining cases could have been both left or right ventricular hypertrophy, from the clinical findings, and the question cannot be satisfactorily answered as to whether or not the right-sided hypertrophy relatively predominated. But, again, it is remarkable that the two sides should have hypertrophied so proportionately as to maintain the normal ratio as determined by an instrument so sensitive as to be of "great importance" in the diagnosis of the *earliest* changes on either side.

One hundred and twenty-five cases showed the sequence of derivations that has come to be associated with left ventricular hypertrophy. Many of these would have been expected from clinical findings.

S. R., aged 66, was admitted to the hospital badly decompensated six times with the diagnosis of arteriosclerosis, myocardial insufficiency, chronic nephritis and dilated aortic arch. Clinically his heart was greatly enlarged to the left and the electrocardiogram (Fig. 12) showed left ventricular hypertrophy, a condition substantiated at necropsy.

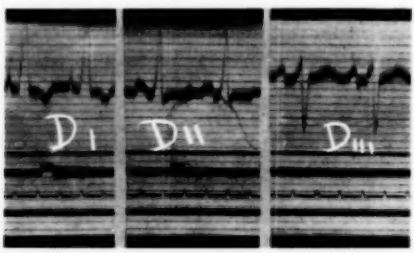


Fig. 12.—Left ventricular hypertrophy according to the electrocardiogram, in spite of the time interval which would have permitted right sided hypertrophy of an extreme degree.

This case, which is quite similar to many others of this series, shows further that where there is a primary left-sided hypertrophy with seemingly right-sided dilatation and hypertrophy—even though of sufficient duration to allow the maximal response of the right side—that the right-sided hypertrophy does not necessarily surpass or equal the hypertrophy already on the left. So that in those cases of expected left ventricular hypertrophy where the electrocardiograms were of the normal sequence, it is not proved that the resulting right ventricular hypertrophy caused a balancing of the two sides to allow a normal electrocardiogram.

On the other hand, many of the cases which, according to the electrocardiogram, had left ventricular hypertrophy, showed clinically normal hearts. Realizing how indefinite is the standard in normal

people when there is no apparent cause for hypertrophy, it would seem of little avail to go into the different clinical histories. Of what importance is the knowledge of left ventricular hypertrophy in a clinically pathological case, if the same degree of left ventricular hypertrophy can be found in an apparently normal person? A further point develops from the perusal of these records. Patients with either recent or long-standing left ventricular hypertrophies did not show as typical a picture of left ventricular hypertrophy as did other patients with apparently normal hearts. Thus, several patients with hearts of normal dimensions from the Roentgen-ray and clinical findings had an inverted $R_2$ and a very long negative $R_3$. Surely there can be no definite relationship between the degree of left ventricular hypertrophy and the degree of inversion of $R_3$.

Another view has been advanced as to the significance of the sequence of derivation. Professor Waller,[1] in several lectures and articles, has suggested that the increase of $R_3$ has to do with a broad angle—the angle made by the electrical axis of the heart with the axis of the body. And he gives examples which show that where the heart is soft and flabby, and, according to the Roentgen ray, is more or less collapsed, mushroom-like over the diagram, the sequence of derivations corresponds to that which Einthoven[2] has associated with left ventricular hypertrophy. While Einthoven's right ventricular hypertrophy is to be connected with the heart that is nearly upright in the body and which is hard and firm, a review of the Roentgen-ray, electrocardiogram and post-mortem findings of this series does not substantiate Waller's conclusions. Thus, several cases with normal electrocardiograms showed, by Roentgen ray, hearts that were practically transverse and which seemed collapsed on the diaphragm. Other cases showed by Roentgen ray the normal position of the heart and right ventricular hypertrophy according to electrocardiogram. Again, one case showed a so-called upright heart with electrocardiographic findings of left ventricular hypertrophy. In this connection an interesting case can be cited: A patient from repeated Roentgen-ray and clinical examinations, was supposed to have dextrocardia. The electrocardiogram showed left ventricular hypertrophy, and was not that associated with transposition of the heart? Autopsy later showed a heart drawn over to the right by pulmonary conditions to the extent necessary to have simulated dextrocardia. The heart was upright in the body and was of fair consistency. This case would, did it substantiate Professor Waller's view, have undoubtedly shown right ventricular hypertrophy.

1. Waller: Lancet, London, 1913, p. 1436.
2. Einthoven: Lancet, London, 1912, i, 853.

Waller, and, following his lead, Pardee,[3] in a recent periodical, substantiate the fact that the electrical axis of the heart makes an angle with the body axis that is calculable easily enough from the different derivations. In no way, however, do they prove the coincidence of the anatomical and electrical axes of the heart, and a review of a number of cases suggests that they do not necessarily agree.

The literature on the electrocardiogram as applied to the diagnosis of cardiac hypertrophy is very confused. Hirschfelder,[4] in his last edition, in speaking of cardiac hypertrophy, says: "The electrocardiograms are, however, characteristic. In hypertrophy of the right ventricle, the R wave is normal or inverted in the first derivation ($D_1$) and large in $D_2$ and $D_3$. In hypertrophy of the left ventricle, the R wave is inverted in $D_2$ and $D_3$." In contradistinction to this dogmatic statement, James and Williams[5] conclude that it is difficult to explain the sequence of derivation obtained in the different types of electrocardiograms. "But the empirically determined fact that a downward deviated $R_1$, with an upward $R_3$, signifies right ventricular hypertrophy is of greatest clinical significance." They continue by saying that in left ventricular hypertrophy, the sequence of derivations is not so useful, because clinical findings are so easy. They seem to regard the electrocardiogram as diagnostic of cardiac hypertrophy.

L. Linitsky[6] says that in young people with low blood-pressure and with small hearts, the I or R wave is relatively small. And he goes on to speak of the effect of age, size of the heart and blood-pressure on the F or T wave.

Einthoven[7] associates right ventricular hypertrophy with a low $R_1$ and high $R_3$, while he diagnoses left ventricular hypertrophy from a relatively low or inverted $R_3$. And his explanation necessitates the coincidence of the electrical and anatomical axes of the heart.

Lewis,[8] in the same way, gives the impression that there is a normal type of electrocardiogram and that deviations from this beyond the age of three months (at which time he states the infant's electrocardiogram changes from the right ventricular hypertrophy type to the normal) are of importance in the diagnosis of cardiac hypertrophy. He very wisely points out the difficulty that often exists in making the diagnosis of cardiac hypertrophy from a physical examination; but he perhaps rather exaggerates the difficulty in concluding that "signs which are

3. Pardee: Jour. Am. Med. Assn., 1914, lxii, 1311.
4. Hirschfelder: Diseases of the Heart and Aorta, Ed. 2, 1913.
5. James and Williams: Med. and Surg. Rep., Presbyterian Hos., 1912.
6. Linitsky: Ztschr. f. Exper. Path. u. Therap., 1911, ix, 669.
7. Einthoven: Arch. f. d. ges. Physiol., 1908, cxxii, 517.
8. Lewis: Brit. Med. Jour., 1912, i, 1423.

customarily employed at the bedside are of little real value in differentiating between right and left ventricular hypertrophy." Certainly the bedside examination is very helpful, and the stress laid on the individual sign "the epigastric pulsation in right ventricular hypertrophy," which Lewis concludes is useless, is a matter of clinical experience; for there are many who have always regarded that sign as of little import. It is the graduation of all the features of a case and their algebraic summation that leads to the clinical diagnosis.

From the pathological study of a series of nine cases, Lewis[9] notes that the clinical discrepancies in clinical and electrocardiographic findings disappear if the exact weights of the two sides of the heart are compared with the electrocardiograms. Such studies are of immense value in the scientific study of the electrocardiogram, but are of little avail in the application to the patient. Even if they are carried out in a sufficient number of cases to confirm the ability of the electrocardiogram to characterize left and right ventricular hypertrophy (a condition not yet obtained), there would be question as to the actual clinical use. It would be hard to say from the electrocardiogram whether the patient was suffering from a mild left-sided lesion with marked right-sided resultants, or whether the lesion was fundamentally a slight right-sided lesion with no trouble on the left side. Without the clinical findings, a diagnosis from the electrocardiogram would seem dangerous, and if the clinical findings differed from the laboratory findings the laboratory findings would have to be corrected. And in addition, there is always the question in a pathological case, "What would the electrocardiogram have shown had the patient been normal?"

As opposed to the views of the English and American observers, the German writers substantiate the opinions expressed in this paper. Hering[10] sums up their conclusions in believing that there is but little dependence to be placed in the electrocardiographic findings in the diagnosis of cardiac hypertrophy.

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With the few cases in this series it is suggested that there is a right ventricular hypertrophy type of electrocardiogram in normal children, which with increasing age gradually passes through the normal to the left ventricular hypertrophy sequence and that there are many exceptions. Attention is called to the absence of a normal cardiovascular system on which to standardize the electrocardiographic findings; especially as age develops, accompanying emphysematous or arteriosclerotic

9. Lewis: Heart, 1914, v, 367.
10. Hering: Deutsch. med. Wchnschr., xxxviii, 2155.

changes appear in varying degrees. The lack of concordance between clinical and electrocardiographic findings is emphasized, and conclusions are pointed out that suggest that the electrocardiogram as it is now understood is of little practical value in the diagnosis of cardiac hypertrophy.

This work was done during the past year, while working in Dr. Thayer's service. It is a pleasure to acknowledge his supervision.

The physical findings were tabulated from the hospital histories of the medical clinic with the permission of Professor Barker.